REPORT DOCUMENTATION PAGE

Form Approved OMB No. 0704-0188

The public reporting burden for this collection of information is estimated to average 1 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information. Send comments regarding this burden estimate or any other aspect of this collection of information, including suggestions for reducing the burden, to the Department of Defense, Executive Service Directorate (0704-0188). Respondents should be aware that notwithstanding any other provision of law, no person shall be subject to any penalty for failing to comply with a collection of information if it does not display a currently valid OMB control number.

			HE ABOVE ORGANIZAT		y valia GIVID of	onto number.
1. REPORT DA	TE (DD-MM-YY	YY) 2. REPC	ORT TYPE			3. DATES COVERED (From - To)
4. TITLE AND S	SUBTITLE	I			5a. CON	ITRACT NUMBER
					5b. GRA	NT NUMBER
					5c. PRO	GRAM ELEMENT NUMBER
6. AUTHOR(S)					5d. PRO	JECT NUMBER
					5e. TAS	K NUMBER
					5f. WOR	K UNIT NUMBER
7. PERFORMIN	IG ORGANIZATI	ON NAME(S) AN	ID ADDRESS(ES)			8. PERFORMING ORGANIZATION REPORT NUMBER
						KEI OKI NOWIDEK
9. Sponsorin	IG/MONITORING	AGENCY NAM	E(S) AND ADDRESS(ES)		10. SPONSOR/MONITOR'S ACRONYM(S)
						11. SPONSOR/MONITOR'S REPORT
						NUMBER(S)
12. DISTRIBUTI	ION/AVAILABILI	TY STATEMENT	•			
13. SUPPLEME	NTARY NOTES					
14. ABSTRACT						
15. SUBJECT T	ERMS					
	CLASSIFICATIO		17. LIMITATION OF ABSTRACT	18. NUMBER OF	19a. NAN	IE OF RESPONSIBLE PERSON
a. REPORT	b. ABSTRACT	C. THIS PAGE	ABOTANO!	PAGES	19b. TEL	EPHONE NUMBER (Include area code)

INSTRUCTIONS FOR COMPLETING SF 298

- **1. REPORT DATE.** Full publication date, including day, month, if available. Must cite at least the year and be Year 2000 compliant, e.g. 30-06-1998; xx-vx-1998.
- **2. REPORT TYPE.** State the type of report, such as final, technical, interim, memorandum, master's thesis, progress, quarterly, research, special, group study, etc.
- **3. DATES COVERED.** Indicate the time during which the work was performed and the report was written, e.g., Jun 1997 Jun 1998; 1-10 Jun 1996; May Nov 1998; Nov 1998.
- **4. TITLE.** Enter title and subtitle with volume number and part number, if applicable. On classified documents, enter the title classification in parentheses.
- **5a. CONTRACT NUMBER.** Enter all contract numbers as they appear in the report, e.g. F33615-86-C-5169.
- **5b. GRANT NUMBER.** Enter all grant numbers as they appear in the report, e.g. AFOSR-82-1234.
- **5c. PROGRAM ELEMENT NUMBER.** Enter all program element numbers as they appear in the report, e.g. 61101A.
- **5d. PROJECT NUMBER.** Enter all project numbers as they appear in the report, e.g. 1F665702D1257; ILIR.
- **5e. TASK NUMBER.** Enter all task numbers as they appear in the report, e.g. 05; RF0330201; T4112.
- **5f. WORK UNIT NUMBER.** Enter all work unit numbers as they appear in the report, e.g. 001; AFAPL30480105.
- **6. AUTHOR(S).** Enter name(s) of person(s) responsible for writing the report, performing the research, or credited with the content of the report. The form of entry is the last name, first name, middle initial, and additional qualifiers separated by commas, e.g. Smith, Richard, J, Jr.
- 7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES). Self-explanatory.

8. PERFORMING ORGANIZATION REPORT NUMBER.

Enter all unique alphanumeric report numbers assigned by the performing organization, e.g. BRL-1234; AFWL-TR-85-4017-Vol-21-PT-2.

- 9. SPONSORING/MONITORING AGENCY NAME(S) AND ADDRESS(ES). Enter the name and address of the organization(s) financially responsible for and monitoring the work.
- **10. SPONSOR/MONITOR'S ACRONYM(S).** Enter, if available, e.g. BRL, ARDEC, NADC.
- **11. SPONSOR/MONITOR'S REPORT NUMBER(S).** Enter report number as assigned by the sponsoring/monitoring agency, if available, e.g. BRL-TR-829; -215.
- **12. DISTRIBUTION/AVAILABILITY STATEMENT.** Use agency-mandated availability statements to indicate the public availability or distribution limitations of the report. If additional limitations/ restrictions or special markings are indicated, follow agency authorization procedures, e.g. RD/FRD, PROPIN, ITAR, etc. Include copyright information.
- **13. SUPPLEMENTARY NOTES.** Enter information not included elsewhere such as: prepared in cooperation with; translation of; report supersedes; old edition number, etc.
- **14. ABSTRACT.** A brief (approximately 200 words) factual summary of the most significant information.
- **15. SUBJECT TERMS.** Key words or phrases identifying major concepts in the report.
- **16. SECURITY CLASSIFICATION.** Enter security classification in accordance with security classification regulations, e.g. U, C, S, etc. If this form contains classified information, stamp classification level on the top and bottom of this page.
- **17. LIMITATION OF ABSTRACT.** This block must be completed to assign a distribution limitation to the abstract. Enter UU (Unclassified Unlimited) or SAR (Same as Report). An entry in this block is necessary if the abstract is to be limited.

A. COVER PAGE

1. Period covered:

Final

2. Title of Proposal:

Extremely high peak power pulsed RF and UWB EMR Effects on Genomic Transcription – Microarray Assessment

3. Grant Number:

AFOSR FA9550-05-1-0021

4. Name of Institution:

University of Texas Health Science Center at San Antonio (UTHSCSA)

5. Author(s) of Report:

Martin L. Meltz, Ph.D., Principal Investigator Bijaya Nayak, Ph.D., Co-Investigator, Principal Investigator – From Dec 1 2007 to Feb 29 2008 Cynthia Galindo, Research Associate

Department of Radiation Oncology University of Texas Health Science Center at San Antonio (UTHSCSA) 7703 Floyd Curl Drive, San Antonio, TX 78250 Office Phone:: 210-567-8025 FAX: 210-567-8051

eMail: navak@uthscsa.edu

Address:

Department of Radiation Oncology Univ. of Texas Health Science Center 7703 Floyd Curl Drive San Antonio, TX 78229-3900

B. Scientific Personnel Supported by this Grant

Martin L. Meltz, Ph.D., Principal Investigator Bijaya Nayak, Ph.D., Co-Investigator and Principal Investigator from Dec 1 2007 to Feb 29 2008

Cynthia Galindo, Research Associate

C. Inventions/Patents/Discoveries

None

D. Collaborations//Consultations/Interactions

In undertaking this research, a natural collaboration developed with two investigators in the Department of Biochemistry, Dr. Susan Weintraub and Mr. Kevin Hakala. These investigators are supported in part to perform proteomic assays on a related AFOSR funded, Multi-University Research Initiative (MURI) Grant Program. The latter grant was issued to Dr. Karl Schoenbach at the Center for Bioelectrics at Old Dominion University in Norfolk, Virginia. Dr. Meltz is the P.I. of the UTHSCSA component of that project. Dr. Schoenbach provided the pulser system for that project to our laboratory. Ms. Galindo, funded on this research project, assists Dr. Meltz in performing the 1 kV/cm UWB GTEMS exposures for this project at the AFRL Laboratory at the former Brooks City-Base, San Antonio, Texas. Ms. Galindo grows and prepares the cells for the exposures, and Dr. Meltz operates the UWB TEMF source. Samples for proteomics are either prepared from the sham and exposed cells by Ms. Galindo for Mr. Hakala, or by Mr. Hakala himself; he performs all of the state of the art proteomics assays under the supervision of Dr. Weintraub.

E. Honors or Awards

None

F. Key Findings

This report will include experiments conducted by the Project Staff of the Radiobiology Group in the Department of Radiation Oncology, UTHSCSA, using the UWB GTEMS source at AFRL, Brooks City-Base. Data from additional selected micro-array experiments performed using the ODU pulser as the EMF sources have been included in this report for comparison purposes.

For all experiments, the cells were exposed to ultrawideband transmitted electromagnetic fields (UWB TEMF) in the GTEM cell at 250 pps for 30 minutes. Temperature data is recorded throughout the entire duration of the experiment at one sample per second, using

Luxtron temperature probe and Labview data collection programming. The temperature is kept at 37°C +/- 0.1°C using a custom designed Watlow airflow temperature regulator and custom designed flask holder incubator boxes. The maximum temperature deviations in each of the UWB exposed and sham exposed flasks were 0.5°C.

F1: Effect of UWB TEMF on Cell Cycle Progression and Apoptosis

Objective:

The effect of UWB TEMEF exposure on cell cycle progression, apoptosis induction, changes in expression of growth arrest and apoptotic genes were examined in different mammalian cells. The cell lines included 244B normal lymphoblastoid cells (p53 competent), MM-6 monocyte cells (p53 compromised), and HL-60 myeloid leukemic cells (p53 null). The UWB TEMF pulses used in this study had an average peak amplitude of 102.17 ± 5.00 kV/m, an average pulse width of 0.80 ± 0.02 ns, and an average rise time of 229.46 ± 25.56 ps. The total exposure time was 90 min, at 250 pulses/sec. The frequency range was from D.C. to greater than 2 GHz.

Methods:

Cell Line Selection

Cell lines with different p53 gene status were selected to address the p53-dependent and p53-independent effects of pulsed UWB TEMF exposures on different genes involved in DNA damage repair, cell growth arrest, and apoptosis. The 244B human lymphoblastoid cells have a wild-type p53 gene activity (p53 competent), the MM-6 human monocyte cells have a compromised p53 gene activity, and the HL-60 human myeloid leukemic cells used are p53-null.

Cell Line Maintenance

The MM-6 cells were maintained in 20 ml of RPMI-1640 medium (Cellgro, Hendron, VA) supplemented with 10% FBS (Hyclone, Logan, UT), 10 mM HEPES (Cellgro), 2mM L-Glutamine (Cellgro), 200U penicillin/200 μ g/ml streptomycin (Cellgro), 2mM non-essential amino acids (NEAA) (Invitrogen, Carlsbad, CA), and 1 vial of OPI (0.15g oxaloacetate, 0,05g pyruvate, and 0.0082g bovine insulin (Sigma, St Louis, MO) per liter of medium. The 244B and HL-60 cells were maintained in 20 ml of RPMI-1640 medium supplemented with 10% heat inactivated FBS, 10 mM HEPES, 2mM L-Glutamine, and 50 μ g/ml gentamicin (Sigma). All of the cells were maintained in disposable T-75 tissue culture flasks with filter caps in a humidified 5% CO₂/95% air atmosphere at 37°C.

Ionizing Radiation Exposure

Cells were exposed to a 5 Gy dose of ¹³⁷Cs gamma rays (Atomic Energy of Canada/Nordion). The dose rate over the course of the experiments ranged from 127.9 cGy/min to 127.2 cGy/min. The cells were exposed at room temperature in T-25 flasks with 10 ml of medium, immediately after removal of the flasks from the 37°C incubator. The flasks were returned to the incubator in the adjoining laboratory immediately after the exposure. The sham controls were carried to and from the irradiation room at the same time.

UWB TEMF Exposure System

The UWB electromagnetic radiation exposures were performed in a custom-built gigatransverse electromagnetic (GTEM) cell originally constructed by Sandia National Laboratories (Albuquerque, NM). The GTEM cell, a tapered two-conductor transmission line with a square cross-section, was positioned so that one ground plane wall was horizontal (8). A modified RG-220 coaxial cable connected the center conductor of the GTEM cell to a source of high voltage. Ionization of nitrogen in a center-conductor gap in the cable generated UWB pulses in the GTEM cell, with the electric field vector directed from the center conductor to the ground conductor. A EG&G ACD(A) D-dot probe mounted in the wall of the GTEM cell was used to detect UWB pulses during the cell exposures. The signal from the probe was sampled with 0.01-ns resolution by a Tektronix SCD 5000 Transient Digitizer. Each digitized wave-form, representing the average of 200 individual pulses, was stored and later processed using a correction algorithm (23) to give the corrected average peak pulse electric field, rise time, and pulse width. The pulses were triggered by an external pulse generator at 250 pulses/s. The mean pulse duration was 0.80 + 0.02 ns. The average peak E-field was 102.17 + 5.00kV/m. The frequencies ranged from D.C. to greater than 2 GHz. The average rise time was 229.46 + 25.56 ps.

Temperature Measurement and Control

Prior to initiating the UWB TEMF exposures, the temperature distribution was determined at the bottom of vertically oriented T-25 flasks positioned in a custom built foam "air incubator" box (described below). The temperature was measured using a Luxtron 3000 Data Acquisition Program. The assessment indicated an average of 37.0 + 0.3°C at the bottom of the flask in the two center position flasks in the foam incubator box. In these two flasks, the temperature distribution around the bottom of the flasks was most uniform. Only these two positions were used for the experimental studies; two dummy flasks filled with 10 ml of RPMI medium were always placed in the two outer positions of the holder. To verify that the temperature remained close to 37°C during and between the intermittent exposures, the temperature in the sham and UWB-exposed flasks were monitored continuously for 5 minutes before, during and for 5 min after the pulsed UWB TEMF exposures (using the Luxtron System). To accomplish this, a Luxtron probe was inserted vertically downward through holes in the foam incubator box and in the caps of each of the two center flasks, to the bottom of the column of the medium in those flasks. The mean temperature during each of the three 30 min UWB exposure periods, and also in the two 30 min UWB off interim periods, was 36.96 + 0.06°C. Two sham exposure flasks were handled in exactly the same manner. These flasks were placed in a second foam holder (with two flasks with medium in the outer positions in the holder) in a Faraday Cage in the anechoic chamber alongside the GTEM cell. The temperature in all the flasks was maintained by computer controlled (Luxtron probe feed back) warm-air flow into the incubator boxes.

Cell Handling for UWB TEMF Exposure

The cells were seeded at 500,000 cells/ml in 10 ml complete growth medium in T-25 flasks with vent caps (Corning) on the day prior to exposure. The cell density was determined by haemocytometer count. Viability was checked using the trypan blue dye exclusion method. The flasks were incubated at 37°C in a humidified 5% CO₂/95% air atmosphere for up to 1 hr, before being transported in a warmed styrofoam container (with 37°C water packs) to a similar incubator in the UWB exposure facility at Brooks City-Base. The flasks were then incubated overnight at 37°C. After checking the performance of the UWB source (pre-exposure check), and 30 min prior to the UWB TEMF exposure, two of the T-25 flasks with cells were placed vertically in the two center positions of the two custom designed 4-flask foam holders (for exposure and sham exposure). The foam holders served as incubators, allowing the temperature of the medium in the flasks to be maintained at 37°C throughout the exposure period. The cells were allowed to settle to the bottom of the flask during this 30 min. The exposure holder was positioned at a distance of 72.7 cm from the opening of the GTEM cell, into which the UWB pulses were emitted. The cells were exposed to UWB pulses intermittently for a total period of 90 minutes (30 min on and 30 min off, repeated three times), at a pulse repetition rate of 250 pulses per second.

Cell Handling Post-Exposure

After the last of the three 30 min UWB TEMF exposures, the flasks were allowed to remain in position in the holder to collect 5 min of post-exposure temperature data. The flasks were then removed from the exposure system and the plug-seal caps were replaced with vented caps. The flasks were placed in the 95% air/5% CO₂ incubator, until the plug-seal caps were replaced and the flasks were transported at near 37°C to the incubator in our laboratory at UTHSCSA. The cells were incubated for the different times indicated below for different assays.

Cell Cycle Analysis

The cell cycle analysis was carried out at 6, 10, and 24 h post-exposure following a standard protocol. Briefly, the cells were harvested, washed with phosphate buffered saline (PBS) and fixed in 70% ethanol. The fixed cells were resuspended in PBS containing propidium iodide (50 μ g/ml), RNase A (500 μ g/ml), and Triton X-100 (0.5%), and incubated in the dark for 1 hour. The cell cycle analysis was performed by flow cytometry in a Becton Dickinson FACSCalibur Flow Cytometer. The results were determined using ModFit software and reported as percentage G0/G1, percentage S, and percentage G2/M cells.

Cell Viability Assay

Cell viability was determined at 2 and 24 hr after UWB TEMF exposures using trypan blue dye exclusion method. A volume of 100 μ l of 0.2% trypan blue was added to 100 μ l of the cell suspension, and the mixture was incubated for 2 minutes. A 10 μ l volume of the cell suspension was loaded onto the hemocytometer slide for counting. The live and dead cells were counted, and the percent viability was determined.

DNA Fragmentation Assay

DNA was isolated from cells using the AquaPure Genomic DNA Isolation Kit (BIORAD, CA) at 6 hour post UWB TEMF exposures. Five μg of DNA was subjected to electrophoresis on a 1% agarose gel, stained with ethidium bromide and visualized by UV illumination. For positive control, cells were treated with staurosporine (Sigma) for 6 hours at a concentration of 0.5 $\mu g/ml$ of medium.

Western Blot Analysis

The p53 protein level and the change in expression of the p53 target gene p21 were examined using Western blot analysis as described earlier (15). Cells were lysed directly in 2X sample buffer {0.125M Tris-Cl, pH 6.8; 4% SDS (Sigma); 0.2M DTT (Sigma) and 0.02% bromphenol blue (Sigma), boiled at 100° C for 10 minutes and centrifuged at 12,000 rpm in a microcentrifuge for 5 minutes to pellet the chromatin. The protein concentration in the supernatant of the whole cell lysate was estimated using the Non-Interfering Protein Assay Kit following manufacturer protocol (Geno Technology Inc., St. Louis, MO). A 50 µg sample of the cell lysate was electrophoresed on a 10% SDSpolyacrylamide gel for ~3 hours and transferred to a nitrocellulose membrane. The membrane was blocked in TBS-T (10 mM Tris-HCl, pH 8; 150 mM NaCl; 0.01% Tween 20) containing 5% blocking agent (Amersham Biosciences, U.K.), and was then probed with the primary antibody (p53, p21, and actin). After washing, the membrane was incubated with horseradish peroxidase-conjugated secondary antibody and was developed with an enhanced chemiluminescence (ECL) kit (Amersham Biosciences). The same membrane was used for all antibodies (p53, p21, and actin) after stripping in strip buffer (125 mM Tris-HCl, pH 6.8; 2% SDS; 0.68% 2-mercaptoethanol). Western blots were quantitated by Densitometry Scanner (Aplha Innotech Corp., CA) using the Fluorchem The fold difference in expression of the genes was determined after software. normalization with the Actin internal control.

RNase Protection Assay (RPA)

Total RNA was isolated at 2, 6, and 24 hr post-exposure using Trizol reagent (Invitrogen, CA). p53 mRNA and transactivation of p53 target genes were examined using the RNase Protection Assay (RPA) as described earlier (15). A human multi-probe template set consisting of p53, p21, PCNA, gadd45, Bax, bcl-2, Bcl-xL, c-fos, and the house keeping genes L32 and GAPDH was used (BDPharmingen, California). The housekeeping genes L32 and GAPDH served as internal controls. The probe set was labeled with T7 RNA polymerase in the presence of α^{32} P-UTP, and was used for RPA as per BDPharmingen's protocol. Briefly, 10 ug of RNA was hybridized with riboprobe (1x10⁶ cpm) in 20 ul of hybridization buffer (40 mM PIPES, pH 6.7; 0.4 M NaCl, 1mM EDTA, and 80% v/v formamide) at 56°C for 12 to 16 hours. Samples were digested with RNase (RNase A and RNase T1 mix) at 30°C for 45 minutes. After proteinase K (Sigma) digestion and phenol-chloroform extraction, the RNA samples were dissolved in 5 µl of loading buffer (80% formamide, 0.1% bromphenol blue, 0.1% xylene cyanol). Samples were electrophoresed for ~1 hour on a 5% denaturing polyacrylamide gel at 50 W in 0.5X TBE. The gels were fixed, dried, and autoradiographed. The gels were scanned using the Typhoon 8600 Variable Mode Imager (Molecular Dynamics, U.K.). Quantitation of the genes was done using the ImageQuant Program (Molecular Dynamics). The fold difference in expression of the genes was determined after normalization with the GAPDH internal control.

Results:

Cell cycle distribution

The UWB TEMF exposures had no effect on the cell cycle progression in 244B cells. There was no difference in the percentage of the UWB TEMF exposed and the sham exposed cells in the different cell cycle stages over the 24 hr observation period (Fig. 1). In positive control cells exposed to 5 Gy dose of ionizing radiation, there was a marginal G2/M arrest (26%), with 65% of cells in G0/G1, and 9% cells in S phase, observed at 24 hr post-exposure (Fig. 1). The 244B cells undergo a G1 arrest in response to phorbol acetate and also undergo a G1/S arrest in response to replication inhibitor hydroxyurea (data not shown). Thus, these cells are responsive to agents which disrupt the cell cycle and cell proliferation. There was also no effect observed on cell cycle distribution in MM-6 and HL-60 cells after UWB TEMF exposures (data not shown).

Figure 1

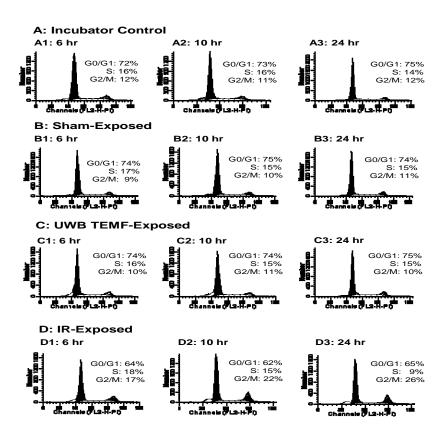


Figure 1: Cell cycle distribution after UWB TEMF exposures in 244B human lymphoblastoid cells. A-Incubator control: A1 (at 6 hour), A2 (at 10 hour), A3 (at 24 hour). B-Sham-exposed: B1 (at 6 hour), B2 (at 10 hour), B3 (at 24 hour). C-UWB TEMF-exposed: C1 (at 6 hour), C2 (at 10 hour), C3 (at 24 hour). D: Ionizing radiation (IR)-exposed (5 Gy): D1 (at 6 hour), D2 (at 10 hour), and D3 (at 24 hour). Left peak, middle plateau, and right peak represent cells in G0/G1, S, and G2/M phases of the cell cycle, respectively.

Cell viability and DNA fragmentation

The cell viability was between 93 to 97% in control, sham, and UWB TEMF-exposed cells in all the three cell lines examined at 2 and 24 hour post-exposure. There was also no DNA fragmentation observed after UWB TEMF exposure in 244B and HL-60 cells at 6 hr post UWB-TEMF exposures (Fig. 2). These observations indicate that there was no induction of cell death within 24 hr after UWB TEMF exposures. In staurosporine treated positive control cells, the majority of the genomic DNA was degraded into oligonucleosomal fragments, resulting in a typical ladder-like pattern (Fig. 2), indicating the onset of apoptotic process. The extent of DNA ladder formation is greater in HL-60 cells than the 244B cells (Fig. 2), showing cell-type specific effects.

Figure 2

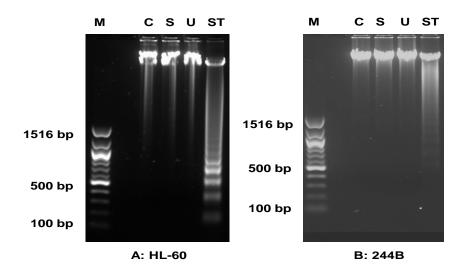


Figure 2: DNA fragmentation analysis after UWB TEMF exposures. A- HL-60, B- 244B cells. C-control, S-sham-exposed, U-UWB TEMF exposed, ST- staurosporine, M-DNA marker.

Expression of growth arrest and apoptotic genes:

Transcriptional activation of p53 and the p53 target genes involved in DNA damage repair (PCNA, and gadd45), cell cycle arrest (p21, gadd45), and apoptosis (bcl-2, bcl-xL, and Bax) was examined after UWB TEMF and 5 Gy ionizing radiation exposures using the RNase protection assay. Representative results for the three cell lines, selected from three independent experiments for each cell line, are shown in figure 3 (244B cells), figure 4 (MM-6 cells), and figure 5 (HL-60 cells). After the UWB TEMF exposures, there was no evidence of transcriptional induction of p53 or the p53 responsive genes p21, gadd45, PCNA, and Bax in 244B (Fig. 3A) or MM-6 (Fig. 4A) cells. In the p53 null HL-60 cells, a change in expression of p21, gadd45, Bax, and PCNA was also not observed in response to UWB TEMF exposure (Fig. 5A), indicating the absence of a p53independent response. In the positive control 244B cells exposed to ionizing radiation, an induction of the p53 target genes p21 (~2.1 fold increase at 2 hr, ~2.2 fold increase at 6 hr and ~1.8 fold increase at 24 hr), gadd45 (~1.5 fold increase at 2 hr, ~1.5 fold increase at 6 hr and ~1.3 fold increase at 24 hr) and PCNA (~1.8 fold increase at 2 hr and ~2 fold increase at 6 hr) was observed, indicating that cell cycle arrest and apoptosis after this type of irradiation could be expected. In the MM-6 (p53 compromised) and HL-60 (p53 null) cells, no change in expression of the p21, gadd45, Bax, bcl-2, bcl-xL, and PCNA genes was observed after exposure to ionizing radiation (Fig. 4A and Fig. 5A), as would be expected for p53 compromised or p53 null cells.

p53 protein Stabilization (Accumulation)

Post-translational modification can block degradation of the p53 protein and result in an increased half life of the protein. As a result, p53 protein would accumulate in the cell; resulting in stabilization of the protein. The level of the p53 protein was examined using Western blot analysis. A representative set of results for 244B and MM-6 cells, out of three independent experiments for each cell line, is shown in Figures 4 and 5. The p53 protein was not accumulated after the UWB TEMF exposure of 244B cells i.e., there was no increase in the p53 protein level as compared to the sham and incubator controls (Fig. 3B). There was also no change in the p53 regulated p21 protein in UWB TEMF exposed 244B cells (Fig. 3B). Increases in p53 and p21 protein levels, and therefore stabilization, were evident after 5 Gy ionizing radiation exposures (Fig. 3B and 4C). The p53 protein level was increased ~1.5 fold at 2 hr and ~1.2 fold at 24 hr post exposure (Fig. 3B and Fig. 4C). The p21 protein level was increased ~1.5 fold at 6 hr and ~1.8 fold at 24 hour post-exposure (Fig. 3B and Fig. 4C). The slight increase in the p53 and p21 protein level in 244B cells after ionizing radiation exposure might be specific to this cell type. In the p53 compromised MM-6 cells, the p53 protein was also not accumulated in response to UWB TEMF exposure (Fig. 4B). The p53 protein level relative to the actin housekeeping gene in both the 244B cells and the MM-6 cells was the same for control, sham-exposed, and UWB TEMF-exposed cells (Fig. 3B and Fig. 4B).

In the p53 compromised MM-6 cells, in contrast to the 244B cells, there was no evidence of p53 accumulation after the 5 Gy ionizing radiation exposures (Fig. 4B). The p53 protein is maintained at higher level in MM-6 cells as compared with the 244B cells (Fig. 4C), indicative of mutant p53 in MM-6 cells. The p21 protein could not be detected in MM-6 cells using western blot analysis (Fig. 4C), indicating a compromised p53 function in these cells. The p21 protein level in p53 null HL-60 cells was maintained at a

lower level, and this level was not altered after the UWB TEMF exposure (Fig. 5B), indicating the absence of a p53 independent effect.

.

Figure 3

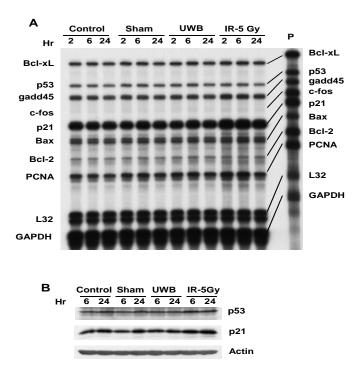


Figure 3: p53 protein accumulation and transactivation of p53 responsive genes in response to pulsed UWB TEMF exposures in human lymphoblastoid 244B cells. A- RNase protection assay (RPA) showing transcription of the p53 and its target genes at 2, 6, and 24 hours post-exposure. B- Western blot analysis showing accumulation of the p53 and p21 protein at 6, and 24 hours post-exposure. Control-incubator control, Sham-sham exposed, UWB-UWB TEMF exposed, IR-5-Gy-ionizing radiation exposed, P-Free probe

.

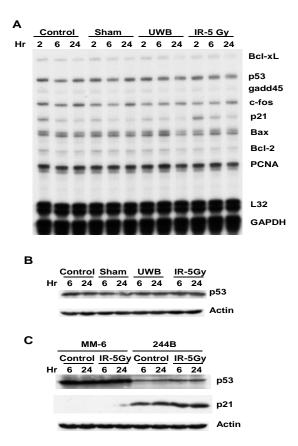


Figure 4: p53 protein accumulation and transactivation of p53 responsive genes in response to pulsed UWB TEMF exposures in human monocyte MM-6 cells. A- RNase protection assay (RPA) showing transcription of the p53 and its target genes at 2, 6, and 24 hours post-exposure. B- Western blot analysis showing accumulation of the p53 protein at 6, and 24 hours post-exposure. C- Western blot analysis showing p53 and p21 protein levels in MM-6 and 244B cells at 6 and 24 hour post-exposure. Control-incubator control, Sham-sham exposed, UWB-UWB TEMF exposed, IR-5-Gy-ionizing radiation exposed.

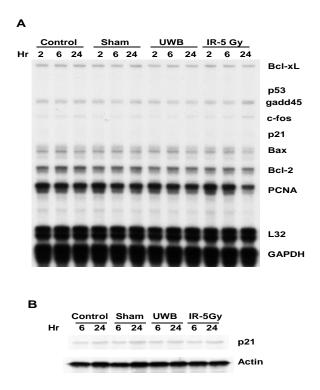


Figure 5: Transactivation of p53 target genes and p21 protein accumulation in response to pulsed UWB TEMF exposures in human myeloid leukemic HL-60 cells. RNase protection assay (RPA) showing transcription of p53 target genes at 2, 6, and 24 hours post-exposure. Control-incubator control, Sham-sham exposed, UWB- UWB TEMF exposed, IR-5-Gy-ionizing radiation exposed. B- Western blot analysis showing p53 and p21 protein levels at 6, and 24 hours post-exposure. Control-incubator control, Sham-sham exposed, UWB-UWB TEMF exposed, IR-5-Gy-ionizing radiation exposed

Conclusion

In this study, the pulsed UWB TEMF exposures were to a very high average peak electric field amplitude ($102.17 \pm 5.10 \text{ kV/m}$), a very rapid rise time ($229.46 \pm 25.56 \text{ ps}$), a very small pulse width ($0.80 \pm 0.02 \text{ ns}$), and a reasonable experimental exposure duration (a total of 90 minutes pulsed UWB TEMF exposure). This study demonstrates for the first time that pulsed UWB electromagnetic field exposure does not induce p53 mRNA expression, does not effect p53 protein accumulation, has no effect on the transactivation of its target genes, and there is no cell growth arrest or apoptosis. Lack of p53 protein accumulation and absence of transcriptional activation of p53 target genes in response to pulsed UWB electromagnetic field exposure suggest that UWB TEMF exposure does not induce DNA damage, and that this type of radiation might not be a stress factor to the cells.

F2: MicroArray Analysis using GeneSpring Software of 244B cells exposed to UWB and 10nsPEF

Objective:

244B cells are exposed to UWB pulses at rate of 250 pulses per second at peak power of 1kV/cm for 30 minutes continuously; 25 pulses at 200kV/cm at rate of 1.5 pps; 10 pulses at 200kV/cm at rate of 1.5pps and 3 pulses at 200kV/cm at rate of 1.5pps. RNA from cells extracted at 2hr and 24 hr post exposure was hybridized onto Affymetrix HGU 133A chips and the resulting data analyzed using GeneSpring analysis software.

Method:

Cells were harvested and RNA was isolated at 2hr and 24hr post each of these exposures. A total of 12 samples were run per experiment because the greater number of replicates will greatly decrease likelihood of false positives.

Intact RNA was assessed using Agilent 2100 BioAnalzyer and the integrity values for all RNA samples were greater than 8.0 (maximum value 10.0). The RNA was then hybridized onto the Affymetrix HGU133-A chips containing 22,000 genes. The intensity of bound dye is measured in an argon laser confocal scanner. The probe arrays are scanned twice and the stored images are aligned and analyzed using the GeneChip software MAS 5.0 (Affymetrix). Signal intensities for each oligonucleotide or Expressed Sequence Tag (EST) are calculated as the difference of filtered perfect match (PM) probes minus mismatch (MM) probes. Signal intensities are globally normalized (using data from all probes on the chip) and scaled to a common mean value according to the Affymetrix protocol. .CHP files provided by the UTHSCSA microarray core facility are imported into GeneSpring and normalized using RMA Preprocessor. The normalized values for the all sham samples (one time point) and each of the RF exposed samples at each experiment time point are averaged together and the mean expression values are used for analysis.

The log of ratio data interpretation is used and the list of genes is filtered using quality controls of filtering on flags, flags must be present or marginal in 3 of 6 samples. The gene list is further reduced by selecting a 2 fold greater than or less than criteria as a ratio between the sham exposed and RF exposed samples. A p value of <0.05 is considered statistically significant.

Results:

Presented below is a "Summary Table of Number of Genes Increasing/Decreasing compared to Sham Controls at Each Time Point for Each Exposure Condition (GeneSpring 7.3 analysis)." As indicated in the methods section, the data in the table below is the average from three independent samples. Each of these samples resulted from an independent exposure on a different exposure day.

Number of Genes Increasing/Decreasing compared to Sham Controls at Each Time Point for Each Exposure Condition (GeneSpring 7.3 analysis).

2 Hour	1 kV/cm UWB	200 kV/cm 3 Pulses	200 kV/cm 10 Pulses	200 kV/cm 25 Pulses	
Increasing	9	39	154	75	
Decreasing	7	16	51	27	
Total	16	55	205	102	
24 Hour	1 kV/cm UWB	200 kV/cm 3 Pulses	200 kV/cm 10 Pulses	200 kV/cm 25 Pulses	
Increasing	10	21	251	42	
Decreasing	3	16	91	53	
Total	13	37	342	95	

Conclusion 1: Differences were observed in the numbers of increases and decreases for the two different time points for each of the four different signals, and differences were also observed in the numbers of increases and decreases for each signal type, with the 10ns PEF signal exposures causing larger numbers of changes.

Below is a list of tables of the specific genes that show changes vs. sham exposed, for each exposure condition and post-exposure assay time. These tables all appear in the appendix provided.

TABLE 1a List of genes increasing at 2 hr post 30 min UWB exposure (1 kV/cm) in pulsed samples compared to sham controls (GeneSpring analysis, 2 fold cutoff)

TABLE 1b List of genes decreasing at 2hr post 30 min UWB exposure (1 kV/cm) in pulsed samples compared to sham controls (GeneSpring analysis, 2 fold cutoff)

TABLE 2a List of genes increasing at 24 hr post 30 min UWB exposure (1 kV/cm) in Pulsed samples compared to sham controls (GeneSpring analysis, 2 fold cutoff)

TABLE 2b List of genes decreasing at 24hr post 30 min UWB exposure (1 kV/cm) in pulsed samples compared to sham controls (GeneSpring analysis, 2 fold cutoff)

TABLE 3a List of Genes Increasing at 2hr post 3 pulse 10ns PEF exposure (200 kV/cm) in pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

TABLE 3b List of Genes Decreasing at 2 hr post 3 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200kV/cm) (GeneSpring analysis, 2 fold

TABLE 4a List of Genes Increasing at 24 hr post 3 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200kV/cm) (GeneSpring analysis, 2 fold cutoff)

TABLE 4b List of Genes Decreasing at 24 hr post 3 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200kV/cm) (GeneSpring analysis, 2 fold

TABLE 5a

List of Genes Increasing at 2 hr post 10 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200kV/cm) (GeneSpring analysis, 2 fold cutoff)

TABLE 5b List of Genes Decreasing at 2 hr post 10 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

TABLE 6a List of Genes Increasing at 24 hr post 10 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

TABLE 6b List of Genes Decreasing at 24 hr post 10 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

TABLE 7a List of Genes Increasing at 2hr post 25 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

TABLE 7b List of Genes Decreasing at 2hr post 25 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

TABLE 8a List of Genes Increasing at 24 hr post 25 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

TABLE 8b List of Genes Decreasing at 24 hr post 25 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

Conclusion 2: The focus of this AFOSR project is the determination of molecular changes after exposure of the human cells to the 0.78 ns, 1 kV/cm UWB GTEMS signal. Examination of Tables 1a, 1b, 2a and 2b, for this type of exposure, indicates that there are no overlapping genes showing changers (either increases or decreases) for the different time points of assay.

Protein Alterations by Proteomics (244B cells) following exposure for 30 minutes to 0.78 ns pulse width UWB, 1 kV/cm average peak power (GTEMS)

Methods: Exponentially growing 244B cells were exposed for 30 min to UWB TEMF (1 kV/cm), and subsequently incubated for 2 hr post-exposure. The cells were harvested, the nuclear extract prepared, and the protein was quantified. The amount of 65 μ g of each nuclear extract was isoelectrically focused on a pH 4 to 7 fixed gradient IEF strip (Bio-Rad), followed by SDS-PAGE on a 12% precast acrylamide gel (Bio-Rad).

Results: 2 hr after a 30 min exposure to a total of 450,000 pulses, no changes were observed in protein expression between exposed cells and sham exposed cells in 2DE gels.

H. Transitions/Technology Transfers

None

APPENDIX

TABLE 1a
List of genes increasing at 2 hr post 30 min UWB exposure (1 kV/cm) in pulsed samples compared to sham controls (GeneSpring analysis, 2 fold cutoff)

Gene Name	Change	Common	Description
202820_at	3.031	AHR	go_component: nucleus [goid 0005634] [evidence IDA] [pmid 10395741];
213036_x_at	2.096	SERCA3	Homo sapiens SERCA3 gene, exons 1-7 (and joined CDS).
218630_at	2.087	FLJ20345	Homo sapiens hypothetical protein FLJ20345 (FLJ20345), mRNA.
200643_at	2.063	HDLBP	synonyms: HBP, VGL; vigilin; go_component: nucleus [goid 0005634] [evidence IEA];
205527_s_at	2.045	GEMIN4	synonyms: HHRF-1, DKFZP434B131, DKFZP434D174; go_component: cytoplasm
216537_s_at	2.042	airm	Homo sapiens mRNA for QA79 membrane protein, splice product airm-1.
218841_at	2.035	ASB8	synonyms: MGC5540, PP14212, FLJ21255; go_process: intracellular signaling cascade synonyms: IB2, JIP2, PRKM8IPL; isoform 1 is encoded by transcript variant 1; PRKM8 interacting protein
205050_s_at	2.013	MAPK8IP2	protein 2;
210715_s_at	2.003	kop	KOP; contains putative transmembrane domain; similar to human placental bikunin;

TABLE 1b List of genes decreasing at 2hr post 30 min UWB exposure (1 kV/cm) in pulsed samples compared to sham controls (GeneSpring analysis, 2 fold cutoff)

Gene Name	Fold Change	Common	Description
211414_at	0.499	GLS	Homo sapiens glutaminase isoform M precursor, mRNA, complete cds. ALL-1 fused gene from chromosome 10; splice variant; Homo sapiens type V AF10 protein (AF10) mR
216509_x_at	0.488	AF10	cds.
214277_at	0.487	COX11	tc35f05.x1 Soares_total_fetus_Nb2HF8_9w Homo sapiens cDNA clone IMAGE:2066625
209006_s_at	0.47	NPD014	Homo sapiens NPD014 (NPD014) mRNA, complete cds.
206420_at	0.441	IGSF6	synonym: DORA; down-regulated by activation (immunoglobulin superfamily);
206255_at	0.436	BLK	synonym: MGC10442; BLK nonreceptor tyrosine kinase; go_function: ATP binding [goid 0005524]
214106_s_at	0.423	GMDS	wi53h03.x1 NCI_CGAP_Co16 Homo sapiens cDNA clone IMAGE:2394005 3

TABLE 2a List of genes increasing at 24 hr post 30 min UWB exposure (1 kV/cm) in Pulsed samples compared to sham controls (GeneSpring analysis, 2 fold cutoff)

Fold		
Change	Common	Description
2.912	PEX5	synonyms: PXR1, PTS1R; peroxisome receptor 1; go_component: peroxisomal membrane [goid 0005778]
2.259	CKLF	transcription factor; Homo sapiens colon Kruppel-like factor (CKLF) mRNA, complete cds.
2.214	C6orf111	xc29c08.x1 NCI_CGAP_Co18 Homo sapiens cDNA clone IMAGE:2585678 3', mRNA sequence. Homo sapiens hypothetical protein MGC12760, mRNA (cDNA clone MGC:12760 IMAGE:4111573), complete
2.17	MGC12760	cds.
2.152	DCN	alternative splice product; Homo sapiens decorin variant C mRNA, complete cds.
2.117	PKNOX2	HUM408A08B Human fetal brain (TFujiwara) Homo sapiens cDNA clone GEN-408A08 5', mRNA sequence.
2.076	SEC23B	
2.048	AP1B1	synonyms: ADTB1, BAM22, AP105A, CLAPB2; isoform a is encoded by transcript variant 1;
2.031	SIX2	go_component: nucleus [goid 0005634] [evidence IEA]; go_function: transcription factor activity [goid 0003700]
2.008	CENPF	350 kDa nuclear phosphoprotein; Human mitosin mRNA, complete cds.

TABLE 2b

List of genes decreasing at 24hr post 30 min UWB exposure (1 kV/cm) in pulsed samples compared to sham controls (GeneSpring analysis, 2 fold cutoff)

Fola		
Change	Common	Description
0.417	HSPH1	602419382F1 NIH_MGC_93 Homo sapiens cDNA clone IMAGE:4526180 5', mRNA sequence.
0.307	RPS11	602155016F1 NIH_MGC_83 Homo sapiens cDNA clone IMAGE:4296050 5', mRNA sequence.

TABLE 3a

List of Genes Increasing at 2hr post 3 pulse 10ns PEF exposure (200 kV/cm) in pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

Gene Name	Fold Change	Gene Symbol	Description
	•	CUL2	cullin 2
203078_at	5.584		
205195_at	4.005	AP1S1	adaptor-related protein complex 1, sigma 1 subunit
222310_at	3.847	SFRS15	splicing factor, arginine/serine-rich 15
201027_s_at	3.64	EIF5B	eukaryotic translation initiation factor 5B phospholipase A2, group VII (platelet-activating factor acetylhydrolase, plasma); phospholipase A2,
206214_at	3.543	PLA2G7	plasma)
218129_s_at	3.318	NFYB	nuclear transcription factor Y, beta
215786_at	3.311		
210042_s_at	3.087	CTSZ	cathepsin Z
201295_s_at	2.914	WSB1	WD repeat and SOCS box-containing 1
210531_at	2.897	NR2C1	nuclear receptor subfamily 2, group C, member 1
208892_s_at	2.852	DUSP6	dual specificity phosphatase 6
203494_s_at	2.781	PIG8	translokin
204748_at	2.707	PTGS2	prostaglandin-endoperoxide synthase 2 (prostaglandin G/H synthase and cyclooxygenase)
205822_s_at	2.69	HMGCS1	3-hydroxy-3-methylglutaryl-Coenzyme A synthase 1 (soluble)
202667_s_at	2.589	SLC39A7	solute carrier family 39 (zinc transporter), member 7
202303_x_at	2.583	SMARCA5	SWI/SNF related, matrix associated, actin dependent regulator of chromatin, subfamily a, member 5
203117_s_at	2.509	USP52	ubiquitin specific peptidase 52
218723_s_at	2.415	RGC32	response gene to complement 32
204622_x_at	2.399	NR4A2	nuclear receptor subfamily 4, group A, member 2
215281_x_at	2.362	POGZ	pogo transposable element with ZNF domain

205191_at	2.361	RP2	retinitis pigmentosa 2 (X-linked recessive)
217299_s_at	2.361	NBN	nibrin
203589_s_at	2.329	TFDP2	transcription factor Dp-2 (E2F dimerization partner 2)
203552_at	2.302	MAP4K5	mitogen-activated protein kinase kinase kinase sinase 5
218886_at	2.242	PAK1IP1	PAK1 interacting protein 1
205196_s_at	2.202	AP1S1	adaptor-related protein complex 1, sigma 1 subunit
221832_s_at	2.173	LUZP1	leucine zipper protein 1
202840_at	2.154	TAF15	TAF15 RNA polymerase II, TATA box binding protein (TBP)-associated factor, 68kDa
211833_s_at	2.129	BAX	BCL2-associated X protein
218040_at	2.114	PRPF38B	PRP38 pre-mRNA processing factor 38 (yeast) domain containing B
201055_s_at	2.096	HNRPA0	heterogeneous nuclear ribonucleoprotein A0
220954_s_at	2.074	PILRB	paired immunoglobin-like type 2 receptor beta
204103_at	2.071	CCL4	chemokine (C-C motif) ligand 4
66053_at	2.071	HNRPUL2	heterogeneous nuclear ribonucleoprotein U-like 2
219399_at	2.062	LIN7C KTN1 :	lin-7 homolog C (C. elegans)
200915_x_at	2.061	PDIA6	kinectin 1 (kinesin receptor); protein disulfide isomerase family A, member 6
216248_s_at	2.049	NR4A2	nuclear receptor subfamily 4, group A, member 2
208942_s_at	2.048	TLOC1	translocation protein 1
206366_x_at	2.01	XCL2	chemokine (C motif) ligand 2

TABLE 3b List of Genes Decreasing at 2 hr post 3 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200kV/cm) (GeneSpring analysis, 2 fold cutoff)

•	Fold	Gene	
Gene Name	Change	Symbol	Description
216556_x_at	0.498		
217663_at	0.493		Transcribed locus, weakly similar to XP_510104.1 PREDICTED: similar to hypothetical protein FLJ25 troglodytes]
221169_s_at	0.493	HRH4	histamine receptor H4
207608_x_at	0.489	CYP1A2	cytochrome P450, family 1, subfamily A, polypeptide 2
203147_s_at	0.485	TRIM14	tripartite motif-containing 14
220704_at	0.48	ZNFN1A1	zinc finger protein, subfamily 1A, 1 (Ikaros)
207827_x_at	0.477	SNCA	synuclein, alpha (non A4 component of amyloid precursor)
220460_at	0.451	SLCO1C1	solute carrier organic anion transporter family, member 1C1
221684_s_at	0.443	NYX	nyctalopin
221352_at	0.427	HUMBINDC	DNA binding protein for surfactant protein B
202946_s_at	0.422	BTBD3	BTB (POZ) domain containing 3
201382_at	0.419	CACYBP	calcyclin binding protein
217090_at	0.414	ADAM3A	ADAM metallopeptidase domain 3a (cyritestin 1)
220052_s_at	0.374	TINF2	TERF1 (TRF1)-interacting nuclear factor 2
217566_s_at	0.334	TGM4	transglutaminase 4 (prostate)
204940_at	0.239	PLN	phospholamban

TABLE 4a

List of Genes Increasing at 24 hr post 3 pulse 10ns PEF exposure ($200\,kV/cm$) in Pulsed samples compared to sham controls (200kV/cm) (GeneSpring analysis, 2 fold cutoff)

Fold Change Common Description

2.912	PEX5	synonyms: PXR1, PTS1R; peroxisome receptor 1; go_component: peroxisomal membrane [goid 0005778]
2.259	CKLF	transcription factor; Homo sapiens colon Kruppel-like factor (CKLF) mRNA, complete cds.
2.214	C6orf111	xc29c08.x1 NCI_CGAP_Co18 Homo sapiens cDNA clone IMAGE:2585678 3', mRNA sequence. Homo sapiens hypothetical protein MGC12760, mRNA (cDNA clone MGC:12760 IMAGE:4111573), complete
2.17	MGC12760	cds.
2.152	DCN	alternative splice product; Homo sapiens decorin variant C mRNA, complete cds.
2.117	PKNOX2	HUM408A08B Human fetal brain (TFujiwara) Homo sapiens cDNA clone GEN-408A08 5', mRNA sequence.
2.076	SEC23B	
2.048	AP1B1	synonyms: ADTB1, BAM22, AP105A, CLAPB2; isoform a is encoded by transcript variant 1;
2.031	SIX2	go_component: nucleus [goid 0005634] [evidence IEA]; go_function: transcription factor activity [goid 0003700]
2.008	CENPF	350 kDa nuclear phosphoprotein; Human mitosin mRNA, complete cds.

TABLE 4b

List of Genes Decreasing at 24 hr post 3 pulse 10ns PEF exposure ($200 \, kV/cm$) in Pulsed samples compared to sham controls (200kV/cm) (GeneSpring analysis, 2 fold cutoff)

Fold		
Change	Common	Description
0.417	HSPH1	602419382F1 NIH_MGC_93 Homo sapiens cDNA clone IMAGE:4526180 5', mRNA sequence.
0.307	RPS11	602155016F1 NIH_MGC_83 Homo sapiens cDNA clone IMAGE:4296050 5', mRNA sequence.

TABLE 5a List of Genes Increasing at 2 hr post 10 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200kV/cm) (GeneSpring analysis, 2 fold cutoff)

	Fold	Gene	
Gene Name	Change	Symbol	Description
209189_at	129.3	FOS	v-fos FBJ murine osteosarcoma viral oncogene homolog
207768_at	120.5	EGR4	early growth response 4
204621_s_at	98.64	NR4A2	nuclear receptor subfamily 4, group A, member 2
202768_at	81.11	FOSB	FBJ murine osteosarcoma viral oncogene homolog B
216248_s_at	31.21	NR4A2	nuclear receptor subfamily 4, group A, member 2
204622_x_at	21.92	NR4A2	nuclear receptor subfamily 4, group A, member 2
201694_s_at	16.07	EGR1	early growth response 1
218880_at	15.72	FOSL2	FOS-like antigen 2
208961_s_at	15.17	KLF6	Kruppel-like factor 6
201041_s_at	14.41	DUSP1	dual specificity phosphatase 1
202859_x_at	13.64	IL8	interleukin 8
209959_at	12.84	NR4A3	nuclear receptor subfamily 4, group A, member 3
211506_s_at	10.07	IL8	interleukin 8
36711_at	8.913	MAFF	v-maf musculoaponeurotic fibrosarcoma oncogene homolog F (avian)
218723_s_at	8.302	RGC32	response gene to complement 32
209795_at	7.692	CD69	CD69 antigen (p60, early T-cell activation antigen)
202672_s_at	7.505	ATF3	activating transcription factor 3
206115_at	7.452	EGR3	early growth response 3
221841_s_at	5.95	KLF4	Kruppel-like factor 4 (gut)
204748_at	5.754	PTGS2	prostaglandin-endoperoxide synthase 2 (prostaglandin G/H synthase and cyclooxygenase)

202014_at	5.513	PPP1R15A	protein phosphatase 1, regulatory (inhibitor) subunit 15A
201531_at	5.39	ZFP36	zinc finger protein 36, C3H type, homolog (mouse)
206916_x_at	5.171	TAT	tyrosine aminotransferase
41386_i_at	4.917	JMJD3	jumonji domain containing 3
207978_s_at	4.857	NR4A3	nuclear receptor subfamily 4, group A, member 3
202497_x_at	4.853	SLC2A3	solute carrier family 2 (facilitated glucose transporter), member 3
203313_s_at	4.69	TGIF	TGFB-induced factor (TALE family homeobox)
201739_at	4.568	SGK	serum/glucocorticoid regulated kinase
206683_at	4.54	ZNF165	zinc finger protein 165
202388_at	4.252	RGS2	regulator of G-protein signalling 2, 24kDa
203068_at	4.06	KLHL21	kelch-like 21 (Drosophila)
209573_s_at	3.996	C18orf1	chromosome 18 open reading frame 1
201693_s_at	3.894	EGR1	early growth response 1
205193_at	3.649	MAFF	v-maf musculoaponeurotic fibrosarcoma oncogene homolog F (avian)
207442_at	3.648	CSF3	colony stimulating factor 3 (granulocyte)
201464_x_at	3.533	JUN	v-jun sarcoma virus 17 oncogene homolog (avian)
207767_s_at	3.518	EGR4	early growth response 4
213593_s_at	3.507	TRA2A	Transformer-2 alpha
209185_s_at	3.449	IRS2	insulin receptor substrate 2
37028_at	3.42	PPP1R15A	protein phosphatase 1, regulatory (inhibitor) subunit 15A
205249_at	3.383	EGR2	early growth response 2 (Krox-20 homolog, Drosophila)
202988_s_at	3.319	RGS1	regulator of G-protein signalling 1
213146_at	3.302	JMJD3	jumonji domain containing 3
221763_at	3.287	JMJD1C	jumonji domain containing 1C
217294_s_at	3.282	ENO1	enolase 1, (alpha)
41387_r_at	3.266	JMJD3	jumonji domain containing 3
215078_at	3.223	SOD2	superoxide dismutase 2, mitochondrial
204194_at	3.22	BACH1	BTB and CNC homology 1, basic leucine zipper transcription factor 1
204440_at	3.15	CD83	CD83 antigen (activated B lymphocytes, immunoglobulin superfamily)
209211_at	3.134	KLF5	Kruppel-like factor 5 (intestinal)
207996_s_at	3.016	C18orf1	chromosome 18 open reading frame 1
204335_at	2.977	FLJ10374	hypothetical protein FLJ10374
201169_s_at	2.897	BHLHB2	basic helix-loop-helix domain containing, class B, 2
205239_at	2.891	AREG	amphiregulin (schwannoma-derived growth factor)
210534_s_at	2.888	EPPB9	B9 protein
206907_at	2.887	TNFSF9	tumor necrosis factor (ligand) superfamily, member 9
204103_at	2.831	CCL4	chemokine (C-C motif) ligand 4
208078_s_at	2.821	SNF1LK	SNF1-like kinase ; SNF1-like kinase
205027_s_at	2.818	MAP3K8	mitogen-activated protein kinase kinase kinase 8
202398_at	2.815	AP3S2	adaptor-related protein complex 3, sigma 2 subunit
214787_at	2.814	MYCPBP	c-myc promoter binding protein
209272_at	2.811	NAB1	NGFI-A binding protein 1 (EGR1 binding protein 1)
219228_at	2.788	ZNF331	zinc finger protein 331
212225_at	2.75	EIF1	eukaryotic translation initiation factor 1
209184_s_at	2.75	IRS2	insulin receptor substrate 2
202067_s_at	2.747	LDLR	low density lipoprotein receptor (familial hypercholesterolemia)
202464_s_at	2.744	PFKFB3	6-phosphofructo-2-kinase/fructose-2,6-biphosphatase 3
202910_s_at	2.743	CD97	CD97 antigen
207433_at	2.704	IL10	interleukin 10
211980_at	2.644	COL4A1	collagen, type IV, alpha 1
500_at		00=//1	

207113_s_at	2.643	TNF	tumor necrosis factor (TNF superfamily, member 2)
214683_s_at	2.595	CLK1	CDC-like kinase 1
214056_s_ut	2.567	MCL1	Myeloid cell leukemia sequence 1 (BCL2-related)
209574_s_at	2.558	C18orf1	chromosome 18 open reading frame 1
213281_at	2.553	0100111	official to open reading name i
219371_s_at		KLF2	Kruppel-like factor 2 (lung)
201466_s_at	2.525	JUN	v-jun sarcoma virus 17 oncogene homolog (avian)
202499_s_at	2.524	SLC2A3	solute carrier family 2 (facilitated glucose transporter), member 3
208926_at	2.522	NEU1	sialidase 1 (lysosomal sialidase)
204802_at	2.514	RRAD	Ras-related associated with diabetes
216449_x_at	2.511	TRA1	tumor rejection antigen (gp96) 1
204015_s_at	2.511	DUSP4	dual specificity phosphatase 4
218630_at	2.477	FLJ20345	hypothetical protein FLJ20345
206744_s_at		ZMYM5	zinc finger, MYM-type 5
200744_s_at 208763_s_at	2.472	TSC22D3	TSC22 domain family, member 3
204857_at	2.417	MAD1L1	MAD1 mitotic arrest deficient-like 1 (yeast)
217739_s_at	2.417	PBEF1	pre-B-cell colony enhancing factor 1
200798_x_at	2.38	MCL1	myeloid cell leukemia sequence 1 (BCL2-related)
214430_at	2.372	GLA	galactosidase, alpha
201925_s_at	2.366	DAF	decay accelerating factor for complement (CD55, Cromer blood group system)
206507_at	2.355	ZNF96	zinc finger protein 96
36829_at	2.353	PER1	period homolog 1 (Drosophila)
200797_s_at	2.342	MCL1	myeloid cell leukemia sequence 1 (BCL2-related)
209900_s_at	2.338	SLC16A1	solute carrier family 16 (monocarboxylic acid transporters), member 1
218999_at	2.333	FLJ11000	hypothetical protein FLJ11000
214098_at	2.325	KIAA1107	KIAA1107
209050_s_at	2.32	RALGDS	ral guanine nucleotide dissociation stimulator
202081_at	2.315	IER2	immediate early response 2
_ 209339_at	2.302	SIAH2	seven in absentia homolog 2 (Drosophila) ; seven in absentia homolog 2 (Drosophila)
218182_s_at	2.298	CLDN1	claudin 1
212338_at	2.274	MYO1D	myosin ID
204014_at	2.272	DUSP4	dual specificity phosphatase 4
216834_at	2.25	RGS1	regulator of G-protein signalling 1
202146_at	2.244	IFRD1	interferon-related developmental regulator 1
221974_at	2.236	SNRPN	Small nuclear ribonucleoprotein polypeptide N
213353_at	2.233	ABCA5	ATP-binding cassette, sub-family A (ABC1), member 5
213568_at	2.233	OSR2	odd-skipped related 2 (Drosophila)
220172_at	2.225	FLJ13096	hypothetical protein FLJ13096
203411_s_at	2.216	LMNA	lamin A/C
214244_s_at	2.212	ATP6V0E	ATPase, H+ transporting, lysosomal 9kDa, V0 subunit e
204587_at	2.194	SLC25A14	solute carrier family 25 (mitochondrial carrier, brain), member 14
209118_s_at	2.173	TUBA3	tubulin, alpha 3
210183_x_at	2.171		
220046_s_at	2.17	CCNL1	cyclin L1
221789_x_at	2.166	RHOT2	ras homolog gene family, member T2
202290_at	2.165	PDAP1	PDGFA associated protein 1
204897_at	2.157	PTGER4	prostaglandin E receptor 4 (subtype EP4) prion protein (p27-30) (Creutzfeld-Jakob disease, Gerstmann-Strausler-Scheinker syndrome, fatal fam
201300_s_at	2.156	PRNP	insomnia)
206366_x_at	2.154	XCL2	chemokine (C motif) ligand 2
219681_s_at	2.145	RAB11FIP1	RAB11 family interacting protein 1 (class I)

219336_s_at	2.143	ASCC1	activating signal cointegrator 1 complex subunit 1	
211414_at	2.14	GLS	glutaminase	
217383_at	2.125	PGK1	Phosphoglycerate kinase 1	
204435_at	2.122	NUPL1	nucleoporin like 1	
215338_s_at	2.12	NKTR	natural killer-tumor recognition sequence	
203224_at	2.119	RFK	riboflavin kinase	
1405_i_at	2.113	CCL5	chemokine (C-C motif) ligand 5	
222207_x_at	2.113		CDNA: FLJ20949 fis, clone ADSE01902	
204794_at	2.112	DUSP2	dual specificity phosphatase 2	
203195_s_at	2.085	NUP98	nucleoporin 98kDa	
202779_s_at	2.084	UBE2S	ubiquitin-conjugating enzyme E2S	
222270_at	2.083	SMEK2	KIAA1387 protein	
215501_s_at	2.082	DUSP10	dual specificity phosphatase 10	
208686_s_at	2.08	BRD2	bromodomain containing 2	
200924_s_at	2.07	SLC3A2	solute carrier family 3 (activators of dibasic and neutral amino acid transport), member 2	
220370_s_at	2.065	USP36	ubiquitin specific peptidase 36	
207630_s_at	2.063	CREM	cAMP responsive element modulator	
202241_at	2.054	TRIB1	tribbles homolog 1 (Drosophila)	
209193_at	2.05	PIM1	pim-1 oncogene ; pim-1 oncogene	
214915_at	2.048	FLJ25476	FLJ25476 protein	
204970_s_at	2.047	MAFG	v-maf musculoaponeurotic fibrosarcoma oncogene homolog G (avian)	
209457_at	2.039	DUSP5	dual specificity phosphatase 5	
217756_x_at	2.037	SERF2	small EDRK-rich factor 2	
212099_at	2.035	RHOB	ras homolog gene family, member B	
203679_at	2.034	TMED1	transmembrane emp24 protein transport domain containing 1	
209096_at	2.033	UBE2V2	ubiquitin-conjugating enzyme E2 variant 2	
212086_x_at	2.032	LMNA	lamin A/C	
208622_s_at	2.028	VIL2	villin 2 (ezrin)	
200755_s_at	2.026	CALU	calumenin	
213988_s_at	2.025	SAT	spermidine/spermine N1-acetyltransferase	
21.4567 a at	2.022	XCL1 ; XCL2	champling (C motif) ligand 1 : champling (C motif) ligand 2	
214567_s_at	2.023 2.021	LDLR	chemokine (C motif) ligand 1; chemokine (C motif) ligand 2	
202068_s_at			low density lipoprotein receptor (familial hypercholesterolemia)	
204951_at	2.016	RHOH	ras homolog gene family, member H	
212300_at	2.015	TXLNA	taxilin alpha	
206060_s_at	2.005	PTPN22	protein tyrosine phosphatase, non-receptor type 22 (lymphoid)	
209102_s_at	2.004	HBP1	HMG-box transcription factor 1	

TABLE 5b List of Genes Decreasing at 2 hr post 10 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

	Fold		
Gene Name	Change	Gene Symbol	Description
222196_at	0.499	LOC286434	hypothetical protein LOC286434
222310_at	0.498	SFRS15	splicing factor, arginine/serine-rich 15
201975_at	0.496	RSN	restin (Reed-Steinberg cell-expressed intermediate filament-associated protein)
207338_s_at	0.496	ZNF200	zinc finger protein 200
203106_s_at	0.493	VPS41	vacuolar protein sorting 41 (yeast)
219321_at	0.493	MPP5	membrane protein, palmitoylated 5 (MAGUK p55 subfamily member 5)

205087_at	0.493	RWDD3	RWD domain containing 3
204146_at	0.493	RAD51AP1	RAD51 associated protein 1
209459_s_at	0.49	ABAT	4-aminobutyrate aminotransferase
219158_s_at	0.486	NARG1	NMDA receptor regulated 1
205934_at	0.484	PLCL1	phospholipase C-like 1
205097_at	0.483	SLC26A2	solute carrier family 26 (sulfate transporter), member 2
209829_at	0.483	C6orf32	chromosome 6 open reading frame 32
216377_x_at	0.478	ALPPL2	alkaline phosphatase, placental-like 2
206809_s_at	0.478	HNRPA3	heterogeneous nuclear ribonucleoprotein A3
209404_s_at	0.476	TMED7	transmembrane emp24 protein transport domain containing 7
204244_s_at	0.472	ASK	activator of S phase kinase
219696_at	0.471	FLJ20054	hypothetical protein FLJ20054
219230_at	0.469	FLJ10970	hypothetical protein FLJ10970
217506_at	0.469		Hypothetical gene supported by BC041875; BX648984
218411_s_at	0.461	MBIP	MAP3K12 binding inhibitory protein 1
206188_at	0.46	ZNF623	zinc finger protein 623
205521_at	0.459	ENDOGL1	Endonuclease G-like 1
202906_s_at	0.452	NBN	nibrin
207711_at	0.45	KIAA0889	KIAA0889 protein
221144_at	0.449		
206163_at	0.44	MAB21L1	mab-21-like 1 (C. elegans)
222266_at	0.426	C19orf2	Chromosome 19 open reading frame 2
209425_at	0.425	AMACR	alpha-methylacyl-CoA racemase
207440_at	0.416	SLC35A2	solute carrier family 35 (UDP-galactose transporter), member A2
222344_at	0.408	C5orf13	Chromosome 5 open reading frame 13
203759_at	0.404	ST3GAL4	ST3 beta-galactoside alpha-2,3-sialyltransferase 4
206346_at	0.403	PRLR	prolactin receptor
206440_at	0.39	LIN7A	lin-7 homolog A (C. elegans)
212637_s_at	0.382	WWP1	WW domain containing E3 ubiquitin protein ligase 1
218986_s_at	0.377	FLJ20035	hypothetical protein FLJ20035
219571_s_at	0.373	ZNF12	zinc finger protein 12
221014_s_at	0.372	RAB33B	RAB33B, member RAS oncogene family; RAB33B, member RAS oncogene family
205300_s_at	0.37	U1SNRNPBP	U11/U12 snRNP 35K
220728_at	0.349		
218689_at	0.342	FANCF	Fanconi anemia, complementation group F
220137_at	0.341	FLJ20674	hypothetical protein FLJ20674
210985_s_at	0.331	SP100	nuclear antigen Sp100
220391_at	0.33	ZBTB3	zinc finger and BTB domain containing 3
220502_s_at	0.314	SLC13A1	solute carrier family 13 (sodium/sulfate symporters), member 1
205091_x_at	0.305	RECQL	RecQ protein-like (DNA helicase Q1-like)
217552_x_at	0.291	CR1	complement component (3b/4b) receptor 1, including Knops blood group system
220295_x_at	0.285	DEPDC1	DEP domain containing 1
215112_x_at	0.171	MCF2L2	MCF.2 cell line derived transforming sequence-like 2

TABLE 6a

List of Genes Increasing at 24 hr post 10 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

Gene Name	Fold	Gene Symbol	Description
-----------	------	-------------	-------------

	Change		
209023_s_at	7.034	STAG2	stromal antigen 2
202514_at	6.403	DLG1	discs, large homolog 1 (Drosophila)
222201_s_at	5.7	CASP8AP2	CASP8 associated protein 2
206188_at	5.592	ZNF623	zinc finger protein 623
215224_at	4.293	RPL23	ribosomal protein L23
209512_at	4.218	HSDL2	hydroxysteroid dehydrogenase like 2
201027_s_at	4.107	EIF5B	eukaryotic translation initiation factor 5B
214658_at	3.979	TMED7	transmembrane emp24 protein transport domain containing 7
212467_at	3.679	DNAJC13	DnaJ (Hsp40) homolog, subfamily C, member 13
205296_at	3.631	RBL1	retinoblastoma-like 1 (p107)
208744_x_at	3.495	HSPH1	heat shock 105kDa/110kDa protein 1
216449_x_at	3.485	TRA1	tumor rejection antigen (gp96) 1
214683_s_at	3.361	CLK1	CDC-like kinase 1
219274_at	3.321	TSPAN12	tetraspanin 12
203429_s_at	3.235	C1orf9	chromosome 1 open reading frame 9
201825_s_at	3.198	SCCPDH	saccharopine dehydrogenase (putative)
219028_at	3.173	HIPK2	homeodomain interacting protein kinase 2
210802_s_at	3.137	HSA9761	dimethyladenosine transferase
209896_s_at	3.112	PTPN11	protein tyrosine phosphatase, non-receptor type 11 (Noonan syndrome 1)
212594_at	3.101	PDCD4	programmed cell death 4 (neoplastic transformation inhibitor)
200791_s_at	3.081	IQGAP1	IQ motif containing GTPase activating protein 1
37079_at	3.078	YDD19	YDD19 protein
212714_at	3.068	LARP4	La ribonucleoprotein domain family, member 4
201711_x_at	3.05	RANBP2	RAN binding protein 2
221428_s_at	3.048	TBL1XR1	transducin (beta)-like 1X-linked receptor 1; transducin (beta)-like 1X-linked recep
202906_s_at	2.98	NBN	nibrin
211352_s_at	2.98	NCOA3	nuclear receptor coactivator 3
221540_x_at	2.977	GTF2H2	general transcription factor IIH, polypeptide 2, 44kDa
222103_at	2.946	ATF1	Activating transcription factor 1
217823_s_at	2.922	UBE2J1	ubiquitin-conjugating enzyme E2, J1 (UBC6 homolog, yeast)
209967_s_at AFFX-	2.917	CREM	cAMP responsive element modulator
HUMISGF3A/M97935_5_at	2.884	STAT1	signal transducer and activator of transcription 1, 91kDa
201830_s_at	2.841	NET1	neuroepithelial cell transforming gene 1
201263_at	2.838	TARS	threonyl-tRNA synthetase
200806_s_at	2.828	HSPD1	heat shock 60kDa protein 1 (chaperonin)
218036_x_at	2.82	NMD3	NMD3 homolog (S. cerevisiae)
201663_s_at	2.817	SMC4L1	SMC4 structural maintenance of chromosomes 4-like 1 (yeast)
201855_s_at	2.815	ASCIZ	ATM/ATR-Substrate Chk2-Interacting Zn2+-finger protein
203675_at	2.773	NUCB2	nucleobindin 2
212219_at	2.751	PSME4	proteasome (prosome, macropain) activator subunit 4
218133_s_at	2.741	NIF3L1	NIF3 NGG1 interacting factor 3-like 1 (S. pombe)
202171_at	2.698	ZNF161	zinc finger protein 161
205034_at	2.691	CCNE2	cyclin E2
200598_s_at	2.665	TRA1	tumor rejection antigen (gp96) 1
218295_s_at	2.632	NUP50	nucleoporin 50kDa
211150_s_at	2.614	DLAT	dihydrolipoamide S-acetyltransferase (E2 component of pyruvate dehydrogenase
200993_at	2.612	IPO7	importin 7
221449_s_at	2.602	CDA08	T-cell immunomodulatory protein ; T-cell immunomodulatory protein
218888_s_at	2.596	NETO2	neuropilin (NRP) and tolloid (TLL)-like 2

212982_at	2.579	ZDHHC17	zinc finger, DHHC-type containing 17
201780_s_at	2.575	RNF13	ring finger protein 13
219326_s_at	2.55	B3GNT1	UDP-GlcNAc:betaGal beta-1,3-N-acetylglucosaminyltransferase 1
210356_x_at	2.539	MS4A1	membrane-spanning 4-domains, subfamily A, member 1
219158_s_at	2.533	NARG1	NMDA receptor regulated 1
218041_x_at	2.526	SLC38A2	solute carrier family 38, member 2
205140_at	2.525	FPGT	fucose-1-phosphate guanylyltransferase
221268_s_at	2.521	SGPP1	sphingosine-1-phosphate phosphatase 1; sphingosine-1-phosphate phosphatase
209096_at	2.511	UBE2V2	ubiquitin-conjugating enzyme E2 variant 2
219109_at	2.499	SPAG16	sperm associated antigen 16
202163_s_at	2.482	CNOT8	CCR4-NOT transcription complex, subunit 8
208852_s_at	2.478	CANX	calnexin
203032_s_at	2.475	FH	fumarate hydratase
207304_at	2.468	ZNF45	zinc finger protein 45
210458_s_at	2.466	TANK	TRAF family member-associated NFKB activator
212515_s_at	2.463	DDX3X	DEAD (Asp-Glu-Ala-Asp) box polypeptide 3, X-linked
205767_at	2.462	EREG	epiregulin
201238_s_at	2.457	CAPZA2	capping protein (actin filament) muscle Z-line, alpha 2
217881_s_at	2.456	CDC27	cell division cycle 27
201946_s_at	2.448	CCT2	chaperonin containing TCP1, subunit 2 (beta)
200605_s_at	2.442	PRKAR1A	protein kinase, cAMP-dependent, regulatory, type I, alpha (tissue specific extingu
214812_s_at	2.442	MOBK1B	MOB1, Mps One Binder kinase activator-like 1B (yeast)
213008_at	2.439	FLJ10719	hypothetical protein FLJ10719
209624_s_at	2.439	MCCC2	methylcrotonoyl-Coenzyme A carboxylase 2 (beta)
211423_s_at	2.438	SC5DL	sterol-C5-desaturase (ERG3 delta-5-desaturase homolog, fungal)-like
213353_at	2.437	ABCA5	ATP-binding cassette, sub-family A (ABC1), member 5
208839_s_at	2.433	CAND1	cullin-associated and neddylation-dissociated 1
213483_at	2.43	PPWD1	peptidylprolyl isomerase domain and WD repeat containing 1
208095_s_at	2.429	SRP72	signal recognition particle 72kDa
211098_x_at	2.427	TMCO1	transmembrane and coiled-coil domains 1
202784_s_at	2.422	NNT	nicotinamide nucleotide transhydrogenase
208786_s_at	2.421	MAP1LC3B	microtubule-associated protein 1 light chain 3 beta
212126_at	2.42	CBX5	Chromobox homolog 5 (HP1 alpha homolog, Drosophila)
221896_s_at	2.417	HIGD1A	HIG1 domain family, member 1A
219166_at	2.409	C14orf104	chromosome 14 open reading frame 104
208097_s_at	2.407	TXNDC	thioredoxin domain containing; thioredoxin domain containing
201153_s_at	2.405	MBNL1	muscleblind-like (Drosophila)
202552_s_at	2.398	CRIM1	cysteine rich transmembrane BMP regulator 1 (chordin-like)
203481_at	2.397	C10orf6	chromosome 10 open reading frame 6
211015_s_at	2.397	HSPA4	heat shock 70kDa protein 4
209248_at	2.393	GHITM	growth hormone inducible transmembrane protein
202691_at	2.39	SNRPD1	small nuclear ribonucleoprotein D1 polypeptide 16kDa
202131_s_at	2.389	RIOK3	RIO kinase 3 (yeast); RIO kinase 3 (yeast)
205345_at	2.385	BARD1	BRCA1 associated RING domain 1
208863_s_at	2.385	SFRS1	splicing factor, arginine/serine-rich 1 (splicing factor 2, alternate splicing factor)
201742_x_at	2.371	SFRS1	splicing factor, arginine/serine-rich 1 (splicing factor 2, alternate splicing factor)
201012_at	2.371	ANXA1	annexin A1
213530_at	2.37	RAB3GAP1	RAB3 GTPase activating protein subunit 1 (catalytic)
204362_at	2.367	SCAP2	src family associated phosphoprotein 2
221568_s_at	2.359	LIN7C	lin-7 homolog C (C. elegans)

217826_s_at	2.351	UBE2J1	ubiquitin-conjugating enzyme E2, J1 (UBC6 homolog, yeast)
212635_at	2.351	TNPO1	Transportin 1
201386_s_at	2.346	DHX15	DEAH (Asp-Glu-Ala-His) box polypeptide 15
36830_at	2.342	MIPEP	mitochondrial intermediate peptidase
219767_s_at	2.34	CRYZL1	crystallin, zeta (quinone reductase)-like 1
217191_x_at	2.337		
204427_s_at	2.324	TMED2	transmembrane emp24 domain trafficking protein 2
203594_at	2.319	RTCD1	RNA terminal phosphate cyclase domain 1
218905_at	2.318	C8orf52	chromosome 8 open reading frame 52
221699_s_at	2.313	DDX50	DEAD (Asp-Glu-Ala-Asp) box polypeptide 50; DEAD (Asp-Glu-Ala-Asp) box poly
201339_s_at	2.313	SCP2	sterol carrier protein 2
221580_s_at	2.312	MGC5306	hypothetical protein MGC5306
217418_x_at	2.312	MS4A1	membrane-spanning 4-domains, subfamily A, member 1
211953_s_at	2.311	RANBP5	RAN binding protein 5
211969_at	2.31	HSPCA	heat shock 90kDa protein 1, alpha
218264_at	2.31	BCCIP	BRCA2 and CDKN1A interacting protein
201975_at	2.308	RSN	restin (Reed-Steinberg cell-expressed intermediate filament-associated protein)
212143_s_at	2.306	IGFBP3	insulin-like growth factor binding protein 3
209861_s_at	2.303	METAP2	methionyl aminopeptidase 2
219974_x_at	2.297	ECHDC1	enoyl Coenzyme A hydratase domain containing 1
220460_at	2.296	SLCO1C1	solute carrier organic anion transporter family, member 1C1
201629_s_at	2.293	ACP1	acid phosphatase 1, soluble
201476_s_at	2.29	RRM1	ribonucleotide reductase M1 polypeptide
211986_at	2.283	AHNAK	AHNAK nucleoprotein (desmoyokin)
218319_at	2.28	PELI1	pellino homolog 1 (Drosophila)
202710_at	2.273	BET1	BET1 homolog (S. cerevisiae)
203654_s_at	2.27	COIL	coilin
221510_s_at	2.266	GLS	glutaminase
217811_at	2.256	SELT	selenoprotein T
213424_at	2.255	KIAA0895	KIAA0895 protein
210439_at	2.255	ICOS	inducible T-cell co-stimulator
201637_s_at	2.252	FXR1	fragile X mental retardation, autosomal homolog 1
214553_s_at	2.242	ARPP-19	cyclic AMP phosphoprotein, 19 kD
221517_s_at	2.24	CRSP6	cofactor required for Sp1 transcriptional activation, subunit 6, 77kDa
201198_s_at	2.239	PSMD1	proteasome (prosome, macropain) 26S subunit, non-ATPase, 1
202527_s_at	2.235	SMAD4	SMAD, mothers against DPP homolog 4 (Drosophila)
200722_s_at	2.229	M11S1	membrane component, chromosome 11, surface marker 1
201486_at	2.228	RCN2	reticulocalbin 2, EF-hand calcium binding domain
211744_s_at	2.227	CD58	CD58 antigen, (lymphocyte function-associated antigen 3); CD58 antigen, (lymph
200692_s_at	2.222	HSPA9B	heat shock 70kDa protein 9B (mortalin-2)
200679_x_at	2.22	HMGB1	high-mobility group box 1
219342_at	2.215	NBLA04196	putative protein product of Nbla04196
200008_s_at	2.214	GDI2	GDP dissociation inhibitor 2; GDP dissociation inhibitor 2
218668_s_at	2.214	RAP2C	RAP2C, member of RAS oncogene family
220199_s_at	2.214	C1orf80	chromosome 1 open reading frame 80
205053_at	2.209	PRIM1	primase, polypeptide 1, 49kDa
214959_s_at	2.202	API5	apoptosis inhibitor 5
201532_at	2.196	PSMA3	proteasome (prosome, macropain) subunit, alpha type, 3
218519_at	2.196	SLC35A5	solute carrier family 35, member A5
208800_at	2.194	SRP72	signal recognition particle 72kDa

209551_at	2.193	YIPF4	Yip1 domain family, member 4
203124_s_at	2.188	SLC11A2	solute carrier family 11 (proton-coupled divalent metal ion transporters), member
219885_at	2.187	FLJ10260	likely ortholog of mouse schlafen 3
203020_at	2.179	RABGAP1L	RAB GTPase activating protein 1-like
209406_at	2.179	BAG2	BCL2-associated athanogene 2
203957_at	2.175	E2F6	E2F transcription factor 6
203893_at	2.166	TAF9	TAF9 RNA polymerase II, TATA box binding protein (TBP)-associated factor, 32k
218247_s_at	2.166	RKHD2 SUB1 ;	ring finger and KH domain containing 2
221727_at	2.157	SUB1P1	SUB1 homolog (S. cerevisiae) ; SUB1 homolog (S. cerevisiae) pseudogene 1
202864_s_at	2.153	SP100	nuclear antigen Sp100
202816_s_at	2.149	SS18	synovial sarcoma translocation, chromosome 18
218723_s_at	2.147	RGC32	response gene to complement 32
218146_at	2.147	GLT8D1	glycosyltransferase 8 domain containing 1
203593_at	2.146	CD2AP	CD2-associated protein
201926_s_at	2.145	DAF	decay accelerating factor for complement (CD55, Cromer blood group system)
217188_s_at	2.142	C14orf1	chromosome 14 open reading frame 1
222037_at	2.14	MCM4	MCM4 minichromosome maintenance deficient 4 (S. cerevisiae)
202420_s_at	2.14	DHX9	DEAH (Asp-Glu-Ala-His) box polypeptide 9
201132_at	2.139	HNRPH2	heterogeneous nuclear ribonucleoprotein H2 (H')
203830_at	2.138	NJMU-R1	protein kinase Njmu-R1
203494_s_at	2.127	PIG8	translokin
205173_x_at	2.127	CD58	CD58 antigen, (lymphocyte function-associated antigen 3)
211968_s_at	2.124	HSPCA	heat shock 90kDa protein 1, alpha
201359_at	2.123	COPB	coatomer protein complex, subunit beta
207830_s_at	2.123	PPP1R8	protein phosphatase 1, regulatory (inhibitor) subunit 8
202850_at	2.119	ABCD3	ATP-binding cassette, sub-family D (ALD), member 3
201581_at	2.117	TXNDC13	thioredoxin domain containing 13
215548_s_at	2.116	SCFD1	sec1 family domain containing 1
220132_s_at	2.116	CLEC2D	C-type lectin domain family 2, member D
215719_x_at	2.115	FAS	Fas (TNF receptor superfamily, member 6)
201970_s_at	2.113	NASP	nuclear autoantigenic sperm protein (histone-binding)
212572_at	2.112	STK38L	serine/threonine kinase 38 like X-ray repair complementing defective repair in Chinese hamster cells 5 (double-s
208643_s_at	2.112	XRCC5	80kDa)
208943_s_at	2.11	TLOC1	translocation protein 1
212006_at	2.106	UBXD2	UBX domain containing 2
207098_s_at	2.103	MFN1	mitofusin 1
209180_at	2.103	RABGGTB	Rab geranylgeranyltransferase, beta subunit
218095_s_at	2.1	TPARL	TPA regulated locus
202200_s_at	2.1	SRPK1	SFRS protein kinase 1
203432_at	2.099	TMPO	thymopoietin
201722_s_at	2.098	GALNT1	UDP-N-acetyl-alpha-D-galactosamine:polypeptide N-acetylgalactosaminyltransfer
200626_s_at	2.097	MATR3	matrin 3
210153_s_at	2.095	ME2	malic enzyme 2, NAD(+)-dependent, mitochondrial
218238_at	2.094	GTPBP4	GTP binding protein 4
204835_at	2.091	POLA	polymerase (DNA directed), alpha
218396_at	2.09	VPS13C	vacuolar protein sorting 13C (yeast)
209200_at	2.085	MEF2C	MADS box transcription enhancer factor 2, polypeptide C (myocyte enhancer factor 2)
209662_at	2.085	CETN3	centrin, EF-hand protein, 3 (CDC31 homolog, yeast)
217640_x_at	2.084	C18orf24	chromosome 18 open reading frame 24
	50 1		

213405_at	2.084	RAB22A	RAB22A, member RAS oncogene family
201018_at	2.084	EIF1AX	eukaryotic translation initiation factor 1A, X-linked
200728_at	2.084	ACTR2	ARP2 actin-related protein 2 homolog (yeast)
204905_s_at	2.083	EEF1E1	eukaryotic translation elongation factor 1 epsilon 1
209754_s_at	2.078	TMPO	thymopoietin
204147_s_at	2.073	TFDP1	transcription factor Dp-1
202651_at	2.073	LPGAT1	lysophosphatidylglycerol acyltransferase 1
209308_s_at	2.072	BNIP2	BCL2/adenovirus E1B 19kDa interacting protein 2
204162_at	2.072	KNTC2	kinetochore associated 2
201514_s_at	2.071	G3BP	Ras-GTPase-activating protein SH3-domain-binding protein
34031_i_at	2.071	KRIT1	KRIT1, ankyrin repeat containing
203497_at	2.07	PPARBP	PPAR binding protein
205684_s_at	2.066	C9orf55	chromosome 9 open reading frame 55
202939_at	2.065	ZMPSTE24	zinc metallopeptidase (STE24 homolog, yeast)
202069_s_at	2.061	IDH3A	isocitrate dehydrogenase 3 (NAD+) alpha
214048_at	2.06	MBD4	methyl-CpG binding domain protein 4
202089_s_at	2.058	SLC39A6	solute carrier family 39 (zinc transporter), member 6
212037_at	2.057	PNN	pinin, desmosome associated protein
212610_at	2.054	PTPN11	protein tyrosine phosphatase, non-receptor type 11 (Noonan syndrome 1)
201832_s_at	2.053	VDP	vesicle docking protein p115
209106_at	2.052	NCOA1	nuclear receptor coactivator 1
212628_at	2.051	PKN2	Protein kinase N2
205437_at	2.048	ZNF211	zinc finger protein 211
202813_at	2.045	TARBP1	TAR (HIV) RNA binding protein 1
209421_at	2.045	MSH2	mutS homolog 2, colon cancer, nonpolyposis type 1 (E. coli)
218179_s_at	2.043	FLJ12716	FLJ12716 protein
222270_at	2.043	SMEK2	KIAA1387 protein
202976_s_at	2.042	RHOBTB3	Rho-related BTB domain containing 3
210568_s_at	2.04	RECQL	RecQ protein-like (DNA helicase Q1-like)
217985_s_at	2.039	BAZ1A	bromodomain adjacent to zinc finger domain, 1A
218911_at	2.037	YEATS4	YEATS domain containing 4
208654_s_at	2.036	CD164	CD164 antigen, sialomucin
201857_at	2.035	ZFR	zinc finger RNA binding protein
218352_at	2.034	RCBTB1	regulator of chromosome condensation (RCC1) and BTB (POZ) domain containing
206555_s_at	2.029	THUMPD1	THUMP domain containing 1
218351_at	2.025	COMMD8	COMM domain containing 8
212330_at	2.023	TFDP1	transcription factor Dp-1
204645_at	2.023	CCNT2	cyclin T2
213470_s_at	2.022	HNRPH1	heterogeneous nuclear ribonucleoprotein H1 (H)
209476_at	2.021	TXNDC	thioredoxin domain containing
213111_at	2.018	PIP5K3	phosphatidylinositol-3-phosphate/phosphatidylinositol 5-kinase, type III
212119_at	2.017	RHOQ	ras homolog gene family, member Q
211945_s_at	2.016	ITGB1	integrin, beta 1 (fibronectin receptor, beta polypeptide, antigen CD29 includes MD
205763_s_at	2.012	DDX18	DEAD (Asp-Glu-Ala-Asp) box polypeptide 18
201084_s_at	2.012	BCLAF1	BCL2-associated transcription factor 1
202268_s_at	2.011	APPBP1	amyloid beta precursor protein binding protein 1
213092_x_at	2.011	DNAJC9	DnaJ (Hsp40) homolog, subfamily C, member 9
212245_at	2.008	MCFD2	multiple coagulation factor deficiency 2
212579_at	2.004	SMCHD1	structural maintenance of chromosomes flexible hinge domain containing 1
212887_at	2.004	SEC23A	Sec23 homolog A (S. cerevisiae)
			- '

TABLE 6b List of Genes Decreasing at 24 hr post 10 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

101a cuto11)			
Gene Name	Fold Change	Gene Symbol	Description
201790_s_at	0.499	DHCR7	7-dehydrocholesterol reductase
216041_x_at	0.499	GRN	granulin
207408_at	0.498	SLC22A14	solute carrier family 22 (organic cation transporter), member 14
214574_x_at	0.496	LST1	leukocyte specific transcript 1
204683_at	0.495	ICAM2	intercellular adhesion molecule 2
214417_s_at	0.494	FETUB	Fetuin B
204340_at	0.494	CXorf12	chromosome X open reading frame 12
217016_x_at	0.493	FLJ23172	hypothetical LOC389177
204265_s_at	0.493	GPSM3	G-protein signalling modulator 3 (AGS3-like, C. elegans)
209050_s_at	0.492	RALGDS	ral guanine nucleotide dissociation stimulator
212430_at	0.492	RNPC1	RNA-binding region (RNP1, RRM) containing 1; RNA-binding region (RNP1, RRM) containing 1
209577_at	0.491	PCYT2	phosphate cytidylyltransferase 2, ethanolamine
209919_x_at	0.491	GGT1	gamma-glutamyltransferase 1
218030_at	0.49	GIT1	G protein-coupled receptor kinase interactor 1
220088_at	0.488	C5R1	complement component 5 receptor 1 (C5a ligand)
209635_at	0.488	AP1S1	adaptor-related protein complex 1, sigma 1 subunit
217782_s_at	0.487	GPS1	G protein pathway suppressor 1
213892_s_at	0.485	APRT	adenine phosphoribosyltransferase
207813_s_at	0.485	FDXR	ferredoxin reductase
218565_at	0.485	C9orf114	chromosome 9 open reading frame 114
207131_x_at	0.485	GGT1	gamma-glutamyltransferase 1
210719_s_at	0.484	HMG20B	high-mobility group 20B
211417_x_at	0.484	GGT1	gamma-glutamyltransferase 1
203828_s_at	0.483	IL32	interleukin 32 ; interleukin 32
208360_s_at	0.482		
50221_at	0.481	TFEB	transcription factor EB
210386_s_at	0.479	MTX1	metaxin 1
206807_s_at	0.479	ADD2	adducin 2 (beta)
211012_s_at	0.477	PML ; LOC161527	promyelocytic leukemia; hypothetical protein LOC161527
218387_s_at	0.476	PGLS	6-phosphogluconolactonase
203370_s_at	0.476	PDLIM7	PDZ and LIM domain 7 (enigma)
202771_at	0.475	FAM38A	family with sequence similarity 38, member A
206560_s_at	0.474	MIA	melanoma inhibitory activity
219247_s_at	0.474	ZDHHC14	zinc finger, DHHC-type containing 14
208284_x_at	0.472	GGT1	gamma-glutamyltransferase 1
209016_s_at	0.472	KRT7	keratin 7
221983_at	0.471	C2orf17	chromosome 2 open reading frame 17
208190_s_at	0.469	LISCH7	liver-specific bHLH-Zip transcription factor
204632_at	0.466	RPS6KA4	ribosomal protein S6 kinase, 90kDa, polypeptide 4
201079_at	0.466	SYNGR2	synaptogyrin 2
218902_at	0.464	NOTCH1	Notch homolog 1, translocation-associated (Drosophila)

201264_at	0.461	COPE	coatomer protein complex, subunit epsilon
34260_at	0.461	KIAA0683	KIAA0683 gene product
203749_s_at	0.461	RARA	retinoic acid receptor, alpha
221549_at	0.46	GRWD1	glutamate-rich WD repeat containing 1
211456_x_at	0.455	RPL35	Similar to 60S ribosomal protein L35
218810_at	0.455	ZC3H12A	zinc finger CCCH-type containing 12A
204789_at	0.452	FMNL1	formin-like 1
203522_at	0.448	CCS	copper chaperone for superoxide dismutase
210001_s_at	0.448	SOCS1	suppressor of cytokine signaling 1
213779_at	0.448	EMID1	EMI domain containing 1
211047_x_at	0.447	AP2S1	adaptor-related protein complex 2, sigma 1 subunit; adaptor-related protein complex 2, sigma 1 s
203085_s_at	0.445	TGFB1	transforming growth factor, beta 1 (Camurati-Engelmann disease)
217756_x_at	0.443	SERF2	small EDRK-rich factor 2
210130_s_at	0.443	TM7SF2	transmembrane 7 superfamily member 2 wingless-type MMTV integration site family, member 5B; wingless-type MMTV integration site family.
221029_s_at	0.443	WNT5B	5B
218208_at	0.44	PQLC1	PQ loop repeat containing 1
215825_at	0.438		Clone 24487 mRNA sequence
202022_at	0.438	ALDOC	aldolase C, fructose-bisphosphate
214041_x_at	0.436	RPL37A	Ribosomal protein L37a
203174_s_at	0.434	ARFRP1	ADP-ribosylation factor related protein 1
202030_at	0.429	BCKDK	branched chain ketoacid dehydrogenase kinase
218376_s_at	0.429	MICAL1	microtubule associated monoxygenase, calponin and LIM domain containing 1
202120_x_at	0.426	AP2S1	adaptor-related protein complex 2, sigma 1 subunit
209414_at	0.422	FZR1	fizzy/cell division cycle 20 related 1 (Drosophila)
211031_s_at	0.421	CYLN2	cytoplasmic linker 2; cytoplasmic linker 2
204857_at	0.418	MAD1L1	MAD1 mitotic arrest deficient-like 1 (yeast)
209215_at	0.415	TETRAN	tetracycline transporter-like protein
208074_s_at	0.414	AP2S1	adaptor-related protein complex 2, sigma 1 subunit
215146_s_at	0.413	KIAA1043	KIAA1043 protein
207163_s_at	0.405	AKT1	v-akt murine thymoma viral oncogene homolog 1
221866_at	0.404	TFEB	transcription factor EB
212625_at	0.403	STX10	syntaxin 10
218548_x_at	0.402	TEX264	testis expressed sequence 264
217133_x_at	0.397	CYP2B6	cytochrome P450, family 2, subfamily B, polypeptide 6
206492_at	0.395	FHIT	fragile histidine triad gene
210183_x_at	0.395		
204889_s_at	0.385	NEURL	neuralized-like (Drosophila)
218130_at	0.38	MGC4368	hypothetical protein MGC4368
200859_x_at	0.368	FLNA	filamin A, alpha (actin binding protein 280)
218083_at	0.361	PTGES2	prostaglandin E synthase 2
211064_at	0.359	LOC115648	similar to hypothetical protein FLJ13659; similar to hypothetical protein FLJ13659
218907_s_at	0.355	MGC3036	hypothetical protein MGC3036
214181_x_at	0.349	LST1	leukocyte specific transcript 1
209083_at	0.349	CORO1A	coronin, actin binding protein, 1A
205213_at	0.348	CENTB1	centaurin, beta 1
201508_at	0.345	IGFBP4	insulin-like growth factor binding protein 4
205212_s_at	0.342	CENTB1	centaurin, beta 1
220565_at	0.34	CCR10	chemokine (C-C motif) receptor 10
209038_s_at	0.34	EHD1	EH-domain containing 1
204164_at	0.33	SIPA1	signal-induced proliferation-associated gene 1

213930_at	0.32		Hypothetical gene supported by AB002449
213746_s_at	0.206	FLNA	filamin A, alpha (actin binding protein 280)

TABLE 7a List of Genes Increasing at 2hr post 25 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

ioia cutoii)	Falsi	0	
Gene Name	Fold Change	Gene Symbol	Description
209189_at	214.3	FOS	v-fos FBJ murine osteosarcoma viral oncogene homolog
207768_at	138.4	EGR4	early growth response 4
202768_at	62.92	FOSB	FBJ murine osteosarcoma viral oncogene homolog B
204621_s_at	37.89	NR4A2	nuclear receptor subfamily 4, group A, member 2
201694_s_at	21.58	EGR1	early growth response 1
209959_at	18.8	NR4A3	nuclear receptor subfamily 4, group A, member 3
208961_s_at	13.59	KLF6	Kruppel-like factor 6
208960_s_at	12.22	KLF6	Kruppel-like factor 6
204622_x_at	12.2	NR4A2	nuclear receptor subfamily 4, group A, member 2
202861_at	10.43	PER1	period homolog 1 (Drosophila)
201041_s_at	10.18	DUSP1	dual specificity phosphatase 1
211506_s_at	10.15	IL8	interleukin 8
202859_x_at	10.08	IL8	interleukin 8
216248_s_at	9.698	NR4A2	nuclear receptor subfamily 4, group A, member 2
218723_s_at	8.215	RGC32	response gene to complement 32
201693_s_at	7.241	EGR1	early growth response 1
36711_at	6.803	MAFF	v-maf musculoaponeurotic fibrosarcoma oncogene homolog F (avian)
209795_at	6.33	CD69	CD69 antigen (p60, early T-cell activation antigen)
201464_x_at	6.124	JUN	v-jun sarcoma virus 17 oncogene homolog (avian)
202672_s_at	5.695	ATF3	activating transcription factor 3
207978_s_at	5.41	NR4A3	nuclear receptor subfamily 4, group A, member 3
206115_at	5.128	EGR3	early growth response 3
218880_at	5.047	FOSL2	FOS-like antigen 2
209967_s_at	5.017	CREM	cAMP responsive element modulator
41386_i_at	4.752	JMJD3	jumonji domain containing 3
213146_at	4.684	JMJD3	jumonji domain containing 3
202014_at	4.422	PPP1R15A	protein phosphatase 1, regulatory (inhibitor) subunit 15A
201531_at	4.387	ZFP36	zinc finger protein 36, C3H type, homolog (mouse)
213593_s_at	4.325	TRA2A	Transformer-2 alpha
202988_s_at	4.232	RGS1	regulator of G-protein signalling 1
37028_at	3.94	PPP1R15A	protein phosphatase 1, regulatory (inhibitor) subunit 15A
202388_at	3.776	RGS2	regulator of G-protein signalling 2, 24kDa
219975_x_at	3.64	THEDC1	thioesterase domain containing 1
207113_s_at	3.346	TNF	tumor necrosis factor (TNF superfamily, member 2)
212569_at	3.296	SMCHD1	structural maintenance of chromosomes flexible hinge domain containing 1
209211_at	3.246	KLF5	Kruppel-like factor 5 (intestinal)
206794_at	3.109	ERBB4	v-erb-a erythroblastic leukemia viral oncogene homolog 4 (avian)
207907_at	3.057	TNFSF14	tumor necrosis factor (ligand) superfamily, member 14
213281_at	3.057		
219228_at	3.021	ZNF331	zinc finger protein 331

220306_at	3.003	FAM46C	family with sequence similarity 46, member C
205249_at	3.002	EGR2	early growth response 2 (Krox-20 homolog, Drosophila)
216322_at	2.902	CD58	CD58 antigen, (lymphocyte function-associated antigen 3)
205510_s_at	2.852	FLJ10038	hypothetical protein FLJ10038
	2.794	KLF4	Kruppel-like factor 4 (gut)
221841_s_at	2.786	CCNL1	cyclin L1
220046_s_at		IER2	·
202081_at	2.762		immediate early response 2
208078_s_at	2.658	SNF1LK	SNF1-like kinase; SNF1-like kinase
207767_s_at	2.639	EGR4	early growth response 4
204243_at	2.634	RLF	rearranged L-myc fusion
204440_at	2.615	CD83	CD83 antigen (activated B lymphocytes, immunoglobulin superfamily)
41387_r_at	2.615	JMJD3	jumonji domain containing 3
206907_at	2.573	TNFSF9	tumor necrosis factor (ligand) superfamily, member 9
203234_at	2.562	UPP1	uridine phosphorylase 1
201466_s_at	2.556	JUN	v-jun sarcoma virus 17 oncogene homolog (avian)
204970_s_at	2.528	MAFG	v-maf musculoaponeurotic fibrosarcoma oncogene homolog G (avian)
216834_at	2.481	RGS1	regulator of G-protein signalling 1
219434_at	2.478	TREM1	triggering receptor expressed on myeloid cells 1
208763_s_at AFFX-r2-Hs28SrRNA-	2.436	TSC22D3	TSC22 domain family, member 3
3_at	2.42	DUIGO	
204794_at	2.399	DUSP2	dual specificity phosphatase 2
204420_at	2.395	FOSL1	FOS-like antigen 1
204103_at	2.383	CCL4	chemokine (C-C motif) ligand 4
AFFX-M27830_5_at	2.376	SOX18	SRY (sex determining region Y)-box 18
212099_at	2.359	RHOB	ras homolog gene family, member B
219681_s_at	2.349	RAB11FIP1	RAB11 family interacting protein 1 (class I)
36829_at	2.347	PER1	period homolog 1 (Drosophila)
209339_at	2.337	SIAH2	seven in absentia homolog 2 (Drosophila); seven in absentia homolog 2 (Drosophila)
209062_x_at	2.326	NCOA3	nuclear receptor coactivator 3
222079_at	2.304		
209573_s_at	2.29	C18orf1	chromosome 18 open reading frame 1
200797_s_at	2.282	MCL1	myeloid cell leukemia sequence 1 (BCL2-related)
209185_s_at	2.267	IRS2	insulin receptor substrate 2
214683_s_at	2.259	CLK1	CDC-like kinase 1
209272_at	2.257	NAB1	NGFI-A binding protein 1 (EGR1 binding protein 1)
203908_at	2.253	SLC4A4	solute carrier family 4, sodium bicarbonate cotransporter, member 4
201169_s_at	2.223	BHLHB2	basic helix-loop-helix domain containing, class B, 2
203313_s_at	2.211	TGIF	TGFB-induced factor (TALE family homeobox)
210413_x_at	2.204	SERPINB4	serpin peptidase inhibitor, clade B (ovalbumin), member 4
213988_s_at	2.201	SAT	spermidine/spermine N1-acetyltransferase
200796_s_at	2.198	MCL1	myeloid cell leukemia sequence 1 (BCL2-related)
204194_at	2.171	BACH1	BTB and CNC homology 1, basic leucine zipper transcription factor 1
212476_at	2.167	CENTB2	centaurin, beta 2
214430_at	2.16	GLA	galactosidase, alpha
213718_at	2.152	RBM4	RNA binding motif protein 4
202648_at	2.147	RPS19	ribosomal protein S19
201739_at	2.143	SGK	serum/glucocorticoid regulated kinase
 219380_x_at	2.14	POLH	polymerase (DNA directed), eta
200798_x_at	2.135	MCL1	myeloid cell leukemia sequence 1 (BCL2-related)
213524_s_at	2.11	G0S2	G0/G1switch 2
=			

202241_at	2.07	TRIB1	tribbles homolog 1 (Drosophila)
202393_s_at	2.064	KLF10	Kruppel-like factor 10
214714_at	2.064	ZNF394	zinc finger protein 394
212381_at	2.056	USP24	ubiquitin specific peptidase 24
219543_at	2.054	MAWBP	MAWD binding protein
204015_s_at	2.051	DUSP4	dual specificity phosphatase 4
203310_at	2.049	STXBP3	syntaxin binding protein 3
219742_at	2.047	PRR7	proline rich 7 (synaptic)
38157_at	2.028	DOM3Z	dom-3 homolog Z (C. elegans)
209184_s_at	2.022	IRS2	insulin receptor substrate 2
204805_s_at	2.017	H1FX	H1 histone family, member X
203752_s_at	2.016	JUND	jun D proto-oncogene

TABLE 7b List of Genes Decreasing at 2hr post 25 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

1014 041011)	Fold		
Gene Name	Change	Gene Symbol	Description
201204_s_at	0.494	RRBP1	Ribosome binding protein 1 homolog 180kDa (dog)
204859_s_at	0.492	APAF1	apoptotic peptidase activating factor
220329_s_at	0.49	C6orf96	chromosome 6 open reading frame 96
203106_s_at	0.487	VPS41	vacuolar protein sorting 41 (yeast)
211479_s_at	0.485	HTR2C	5-hydroxytryptamine (serotonin) receptor 2C
219105_x_at	0.483	ORC6L	origin recognition complex, subunit 6 homolog-like (yeast)
209766_at	0.479	PRDX3	peroxiredoxin 3
202552_s_at	0.478	CRIM1	cysteine rich transmembrane BMP regulator 1 (chordin-like)
205527_s_at	0.472	GEMIN4	gem (nuclear organelle) associated protein 4
205305_at	0.459	FGL1	fibrinogen-like 1
219802_at	0.458	FLJ22028	hypothetical protein FLJ22028
201657_at	0.449	ARL1	ADP-ribosylation factor-like 1
219433_at	0.433	BCOR	BCL6 co-repressor
217558_at	0.428	CYP2C9	cytochrome P450, family 2, subfamily C, polypeptide 9
201975_at	0.427	RSN	restin (Reed-Steinberg cell-expressed intermediate filament-associated protein)
202430_s_at	0.405	PLSCR1	phospholipid scramblase 1
214785_at	0.401	VPS13A	vacuolar protein sorting 13A (yeast)
203165_s_at	0.383	SLC33A1	solute carrier family 33 (acetyl-CoA transporter), member 1
213694_at	0.373	RSBN1	round spermatid basic protein 1
218411_s_at	0.368	MBIP	MAP3K12 binding inhibitory protein 1
213951_s_at	0.361	TBPIP	TBP-1 interacting protein
221540_x_at	0.351	GTF2H2	general transcription factor IIH, polypeptide 2, 44kDa
209910_at	0.35	SLC25A16	solute carrier family 25 (mitochondrial carrier; Graves disease autoantigen), member 16
221561_at	0.349	SOAT1	sterol O-acyltransferase (acyl-Coenzyme A: cholesterol acyltransferase) 1
201295_s_at	0.312	WSB1 COX11;	WD repeat and SOCS box-containing 1 COX11 homolog, cytochrome c oxidase assembly protein (yeast); COX11 homolog, cytochrome c
214277_at	0.18	COX11P	pseudogene
202422_s_at	0.179	ACSL4	acyl-CoA synthetase long-chain family member 4

TABLE 8a List of Genes Increasing at 24 hr post 25 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

•	ioia catori)	Fold		
	Gene Name	Change	Gene Symbol	Description
	203213_at	3.401	CDC2	Cell division cycle 2, G1 to S and G2 to M
	214193_s_at	3.09	C1orf107	chromosome 1 open reading frame 107
	213253_at	3.017	SMC2L1	SMC2 structural maintenance of chromosomes 2-like 1 (yeast)
	203092_at	2.871	TIMM44	translocase of inner mitochondrial membrane 44 homolog (yeast)
	221773_at	2.73	ELK3	ELK3, ETS-domain protein (SRF accessory protein 2)
	205885_s_at	2.722	ITGA4	integrin, alpha 4 (antigen CD49D, alpha 4 subunit of VLA-4 receptor)
	213677_s_at	2.599	PMS1	PMS1 postmeiotic segregation increased 1 (S. cerevisiae)
	218842_at	2.569	FLJ21908	hypothetical protein FLJ21908
	202904_s_at	2.553	LSM5	LSM5 homolog, U6 small nuclear RNA associated (S. cerevisiae)
	210970_s_at	2.519	IBTK	inhibitor of Bruton agammaglobulinemia tyrosine kinase
	221230_s_at	2.444	ARID4B	AT rich interactive domain 4B (RBP1- like) ; AT rich interactive domain 4B (RBP1- like)
	222103_at	2.323	ATF1	Activating transcription factor 1
	218411_s_at	2.257	MBIP	MAP3K12 binding inhibitory protein 1
	203735_x_at	2.253	PPFIBP1	PTPRF interacting protein, binding protein 1 (liprin beta 1)
	218694_at	2.249	ARMCX1	armadillo repeat containing, X-linked 1
	203403_s_at	2.244	RNF6	ring finger protein (C3H2C3 type) 6
	219105_x_at	2.229	ORC6L	origin recognition complex, subunit 6 homolog-like (yeast)
	209803_s_at	2.223	PHLDA2	pleckstrin homology-like domain, family A, member 2
	215978_x_at	2.188	LOC152719	hypothetical protein LOC152719
	203411_s_at	2.174	LMNA	lamin A/C
	202146_at	2.172	IFRD1	interferon-related developmental regulator 1
	217019_at	2.169	RPS4X	ribosomal protein S4, X-linked
	217185_s_at	2.164	ZNF259	zinc finger protein 259
	203049_s_at	2.161	KIAA0372	KIAA0372
	218962_s_at	2.16	FLJ13576	hypothetical protein FLJ13576
	217986_s_at	2.133	BAZ1A	bromodomain adjacent to zinc finger domain, 1A
	204333_s_at	2.123	AGA	aspartylglucosaminidase
	218681_s_at	2.117	SDF2L1	stromal cell-derived factor 2-like 1
	202565_s_at	2.117	SVIL	supervillin
	218053_at	2.111	FNBP3	formin binding protein 3
	217941_s_at	2.099	ERBB2IP	erbb2 interacting protein
	203306_s_at	2.087	SLC35A1	solute carrier family 35 (CMP-sialic acid transporter), member A1
		2.08	C1orf25	chromosome 1 open reading frame 25; chromosome 1 open reading frame 25 ubiquitin-conjugating enzyme E2D 1 (UBC4/5 homolog, yeast); ubiquitin-conjugating enzyme E2D 1 (UBC4/5 homolog, yeast)
	211764_s_at	2.08	UBE2D1	yeast)
	212996_s_at	2.079	C21orf108	chromosome 21 open reading frame 108
	213072_at	2.079	CYHR1	cysteine/histidine-rich 1
	209210_s_at	2.07	PLEKHC1	pleckstrin homology domain containing, family C (with FERM domain) member 1
	206816_s_at	2.064	SPAG8	sperm associated antigen 8
	219289_at	2.03	FLJ20718 HSPA1A:	hypothetical protein FLJ20718
	200800_s_at	2.016	HSPA1A ; HSPA1B	heat shock 70kDa protein 1A; heat shock 70kDa protein 1B
	210766_s_at	2.012	CSE1L	CSE1 chromosome segregation 1-like (yeast)
	218050_at	2.004	UFM1	ubiquitin-fold modifier 1
				•

TABLE 8b List of Genes Decreasing at 24 hr post 25 pulse 10ns PEF exposure (200 kV/cm) in Pulsed samples compared to sham controls (200 kV/cm) (GeneSpring analysis, 2 fold cutoff)

Cone Nome	Fold	Cana Symbol	Description
Gene Name	Change	Gene Symbol	Description
202545_at	0.498	PRKCD	protein kinase C, delta
221011_s_at	0.494	LBH	likely ortholog of mouse limb-bud and heart gene; likely ortholog of mouse limb-bud and heart g
209392_at	0.491	ENPP2	ectonucleotide pyrophosphatase/phosphodiesterase 2 (autotaxin)
206134_at 222088_s_at	0.489 0.487	ADAMDEC1 SLC2A3; SLC2A14	ADAM-like, decysin 1 solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 2 (facilitated glucose transporter), member 3; solute carrier family 3 (facilitated glucose transporter), member 3 (facilitated glucose transporter), member 3; solute carrier family 3 (facilitated glucose transporter), member 3 (facilitated
206760_s_at	0.485	FCER2	Fc fragment of IgE, low affinity II, receptor for (CD23A)
201753_s_at	0.482	ADD3	adducin 3 (gamma)
213349_at	0.481	TMCC1	transmembrane and coiled-coil domain family 1
219472_at	0.472	MGC11266	hypothetical protein MGC11266
214658_at	0.469	TMED7	transmembrane emp24 protein transport domain containing 7
201790_s_at	0.468	DHCR7	7-dehydrocholesterol reductase
201790_s_at	0.467	MARCKS	myristoylated alanine-rich protein kinase C substrate
201070_s_at 211016_x_at	0.467	HSPA4	heat shock 70kDa protein 4
		XCL2	·
206366_x_at	0.464		chemokine (C motif) ligand 2
204025_s_at	0.461	PDCD2	programmed cell death 2
218045_x_at	0.459	PTMS	parathymosin
211330_s_at	0.457	HFE	hemochromatosis
209795_at	0.448	CD69	CD69 antigen (p60, early T-cell activation antigen)
207245_at	0.447	UGT2B17	UDP glucuronosyltransferase 2 family, polypeptide B17
214772_at	0.447	C11orf41	chromosome 11 open reading frame 41
202539_s_at	0.446	HMGCR	3-hydroxy-3-methylglutaryl-Coenzyme A reductase
208325_s_at	0.445	AKAP13	A kinase (PRKA) anchor protein 13
205484_at	0.442	SIT1	signaling threshold regulating transmembrane adaptor 1; signaling threshold regulating transme
214567_s_at	0.439	XCL1; XCL2	chemokine (C motif) ligand 1; chemokine (C motif) ligand 2
202770_s_at	0.433	CCNG2	cyclin G2
212331_at	0.433	RBL2	retinoblastoma-like 2 (p130)
204859_s_at	0.43	APAF1	apoptotic peptidase activating factor
201745_at	0.429	PTK9	PTK9 protein tyrosine kinase 9
201382_at	0.429	CACYBP	calcyclin binding protein
218111_s_at	0.428	CMAS	cytidine monophosphate N-acetylneuraminic acid synthetase
215916_at	0.428	CHRNE	cholinergic receptor, nicotinic, epsilon polypeptide
202131_s_at	0.427	RIOK3	RIO kinase 3 (yeast); RIO kinase 3 (yeast)
213025_at	0.426	THUMPD1	THUMP domain containing 1
221874_at	0.424	KIAA1324	KIAA1324
200832_s_at	0.423	SCD	stearoyl-CoA desaturase (delta-9-desaturase)
207386_at	0.415	CYP7B1	cytochrome P450, family 7, subfamily B, polypeptide 1
212819_at	0.412	ASB1	ankyrin repeat and SOCS box-containing 1
214698_at	0.407	ROD1	ROD1 regulator of differentiation 1 (S. pombe)
201848_s_at	0.389	BNIP3	BCL2/adenovirus E1B 19kDa interacting protein 3
205408_at	0.387	MLLT10	myeloid/lymphoid or mixed-lineage leukemia (trithorax homolog, Drosophila); translocated to, 10
202855_s_at	0.366	SLC16A3	solute carrier family 16 (monocarboxylic acid transporters), member 3
205179_s_at	0.365	ADAM8	ADAM metallopeptidase domain 8 ; ADAM metallopeptidase domain 8
204348_s_at	0.364	AK3L1	adenylate kinase 3-like 1

207002_s_at	0.364	PLAGL1	pleiomorphic adenoma gene-like 1
202430_s_at	0.356	PLSCR1	phospholipid scramblase 1
208671_at	0.348	SERINC1	serine incorporator 1
204567_s_at	0.341	ABCG1	ATP-binding cassette, sub-family G (WHITE), member 1
204821_at	0.339	BTN3A3	butyrophilin, subfamily 3, member A3
201849_at	0.337	BNIP3	BCL2/adenovirus E1B 19kDa interacting protein 3
202022_at	0.312	ALDOC	aldolase C, fructose-bisphosphate
210512_s_at	0.296	VEGF	vascular endothelial growth factor
218149_s_at	0.25	ZNF395	zinc finger protein 395
221478_at	0.238	BNIP3L	BCL2/adenovirus E1B 19kDa interacting protein 3-like; BCL2/adenovirus E1B 19kDa interacting